SUGARBEET RESEARCH

2004 REPORT

FOREWARD

SUGARBEET RESEARCH is an annual compilation of progress reports concerning research by U.S. Department of Agriculture, Agricultural Research Service investigators and other cooperators who are engaged in sugarbeet research. The report was assembled and produced at the expense of the Beet Sugar Development Foundation, and is for the sole use of its members and the cooperators. Much of the data has not been sufficiently confirmed to justify general release and interpretations may be modified with additional experimentation. This report is not intended for publication and should not be used for cited reference nor quoted in publicity or advertising. Reproduction of any portion of the material contained herein will not be permitted without the specific consent of the contributor and the Beet Sugar Development Foundation.

The report presents results of investigations strengthened by contributions received under Cooperative Agreement between the USDA Agricultural Research Service and the Beet Sugar Development Foundation, along with the California Beet Growers Association, the Western Joint Research Committee, and the Sugarbeet and Education Board of Minnesota and North Dakota.

Trade names occur in this report solely to provide specific information and do not signify endorsement by the U.S. Department of Agriculture, the Beet Sugar Development Foundation or any of the cooperating organizations.

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SUGARBEET RESEARCH

2004 REPORT

Section A

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  Western Sugar Growers Research Committee

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VARIETY TRIALS

ABSTRACTS OF PAPERS PUBLISHED OR APPROVED FOR PUBLICATION, 2004

Grube, R.C., W.M. Wintermantel, P. Hand', R. Aburomia, D.A.C. Pink, and E.J. Ryder. 2005. Genetic analysis and mapping of resistance to lettuce dieback, a soilborne disease caused by tombusviruses. *Theoretical and Applied Genetics* **110: 259-268.**

A diverse collection of modern, heirloom and specialty cultivars, Plant Introduction (PI) accessions, and breeding lines of lettuce were screened for susceptibility to lettuce dieback, which is a disease caused by soilborne viruses of the family *Tombusviridae*. Of the 241 genotypes tested in field experiments, 76 remained symptom-free in infested fields and were therefore classified as resistant to dieback. Overall, resistant genotypes were as prevalent among modern cultivars as in heirloom cultivars or primitive germplasm. Within modern germplasm, however, all crisphead (iceberg) cultivars were resistant, while all romaine cultivars were susceptible. Using enzyme-linked immunosorbent assay (ELISA), tombusviruses were detected in leaves of some plants of resistant genotypes that were grown in infested fields or that were inoculated as seedlings in the growth chamber, suggesting that symptom-free plants are not immune to viral infection. The inheritance of resistance was studied for both 'Salinas', a modern iceberg cultivar, and PI 491224, the progenitor of recently released romaine germplasm with resistance to lettuce dieback. Resistance was conferred by a dominant allele at a single locus in both genotypes. The tombusvirus resistance locus from 'Salinas', *Tvr1*, was mapped in an intraspecific *L. sativa* population to a location that corresponds to linkage group 2 on the consensus map of *Lactuca*. The largest cluster of resistance genes in lettuce, the *Dm1/Dm3* cluster, is found on this linkage group, however, the precise position of *Tvr1* relative to this cluster has not yet been determined. To our knowledge, *Tvr1* is the first tombusvirus resistance gene identified for `any plant host`.

Lewellen, R.T. 2004. Registration of rhizomania resistant, monogerm populations C869 and C869CMS sugarbeet. Crop Sci. 44:357-358.

Sugar beet (*Beta vulgaris* L.) population C869 (Reg. no. GP-226, PI628754) and its cytoplasmic male-sterile (CMS) counterpart C869CMS (Reg. no. GP-227, PI628755) were developed by the USDA-ARS in cooperation with the Beet Sugar Development Foundation and the California Beet Growers Association. These lines were released in 2002.

C869 is a monogerm (*mm*), O-type, self-fertile (S^f), genetic-male-sterile ($A\_:aa$) facilitated, random-mated population. It segregates for resistance to rhizomania (caused by *Beet necrotic yellow vein* virus) conditioned by the *Rz1* allele. It has mostly red (*R*) hypocotyls. It is moderately resistant to *Beet curly top virus* (BCTV). C869 has wide variability for reaction to bolting, Erwinia rot (caused by *Erwinia carotovora* subsp. *betavasculorum* Thomson et al.), and powdery mildew (caused by *Erysiphe polygoni* DC.). C869 is an N-type for sucrose concentration with average sugar yield combining ability.

C869CMS is the cytoplasmic male sterile counterpart of C869. It will facilitate rapid development of CMS equivalents of lines extracted or developed from C869. It also may be useful as a monogerm, CMS tester to evaluate multigerm lines for general combining ability.

C869 is a moderately diverse population with good monogerm and O-type traits. It produces vigorous plants and high seed yield. Before 1995, the germplasm base of C869 involved developing and recombining subpopulations and selected progeny lines from sources. Collectively, C869 comprises about 44% of its germplasm from C790 (PI515964) (Lewellen and Skoyen, 1988) through C890 (PI593700) (Lewellen, 1998); 12.5% from C310 (C6) (PI590873) (Lewellen and Skoyen, 1988); 12.5% from curly top and Erwinia resistant monogerm inbred C1546 (PI590649) (McFarlane and Skoyen, 1965); and about 31% from the original source of *Rz1* (Biancardi et al., 2002). C790 was a broad based monogerm, self-fertile population that had undergone five cycles of S_1 progeny recurrent selection for sugar yield and was the source of monogerm inbreds such as C790-15 (PI564758) (Lewellen, 1994). C310 was a monogerm, self-
fertile population that had proven valuable as a source of lettuce infectious yellows virus resistant parental lines, e.g., C301 (PI590717) (Lewellen and Skoyen, 1987). Since 1995 when population 867 [(C310 x C546)aa x Rz source] and C890 were combined to form 5869, the progenitor of C869, four cycles of selection have been done. These included individual and combined selections for monogerm, rhizomania resistance, O-type, resistance to Erwinia, powdery mildew, bolting, and for higher sucrose content. From these cycles of selection, subpopulations 7869NB, 7869, and 8869 were formed. Mother root selections from these were recombined in 1999 to produce 9869. In 2000, high quality, monogerm plants of 9869 were selfed to produce selfed progeny families. These families were indexed for O-type and separately evaluated for resistance to rhizomania. About 600 plants from 24 selfed families (i.e., 24 S_0 plants) that appeared to be O-type and have resistance to rhizomania were recombined through their genetic male sterile segregants to produce 1869. Seed of 1869 is being released as C869. In 1996, plants of 5869 were increased through their male sterile segregants to produce 6869. Population 6869 was not used directly to produce C869 but was made available for genetic research and tentatively called C869 (McGrath et al., 1999).

C869 should be useful as a source of resistance to rhizomania and other diseases in a monogerm, O-type background. Sufficient genetic variability should still occur to permit continued population improvement and as a source of potential parental lines. C869 may be useful also as a base population from which to develop additional populations and breeding lines and from which to develop selfed progeny for mapping molecular markers.

Lewellen, R.T. 2004. Registration of sugarbeet germplasm lines C67/2, C69/2, C78/3, and C80/2 with resistance to virus yellows and rhizomania. Crop Sci. 44:358-359.

Sugarbeet (*Beta vulgaris* L.) germplasm lines C67/2 (Reg. no. GP-229, PI628750), C69/2 (Reg. no. GP-230, PI628751), C78/3 (Reg. no. GC-231, PI628752), and C80/2 (Reg. no. GC-232, PI628753) were developed by the USDA-ARS in cooperation with the Beet Sugar Development Foundation, and the California Beet Growers Association. These lines were released in 2002. These are self-sterile ($S^s S^s$), multigerm *(MM)* lines that segregate for resistance to rhizomania caused by *Beet necrotic yellow vein virus*. Resistance to rhizomania is conditioned by *Rz1*. These lines have predominantly red (*R_*) hypocotyls. Earlier versions of these lines have been released. They

encompass a broad cross section of the "Salinas" multigerm, germplasm base. The origin and development of these breeding lines spans 20 to 60 years of breeding improvements in productivity and combined disease resistance. Sugar yield tends to be primarily of the N-type but full-sib and other types of progeny tests have shown wide genetic variability for components of productivity. Selection pressure has been exerted to improve resistance to virus yellows caused by the *Beet yellows virus* (BYV), *Beet western yellows virus* (BWYV), and *Beet Chlorosis virus* complex; *Erwinia carotovora* subsp. *betavasculorum* Thomson et al.; *Erysiphe polygoni* DC, the cause of powdery mildew; rhizomania; *Peronospora farinosa* (Fr.:Fr.) Fr., the cause of downy mildew; and *Uromyces betae* J. Kickx fil., the cause of rust.

C67/2 was released from C67 (PI599340) in 1998. Since that release, this breeding line has undergone two additional cycles of recurrent phenotypic selection. In both cycles, emphasis was placed on selecting mother roots for sucrose concentration, size, and conformation from field plants grown under rhizomania conditions, inoculated with virus yellows and sugarbeet *Erwinia*, and naturally infected with powdery mildew. Plants that bolted before harvest were eliminated. C67/2 is estimated to have about 10% of its germplasm from *B.vulgaris* subsp. *maritima* (*Bvm*). The *Bvm* germplasm was derived from R322Y3%, a component of C51 (PI593694) (Lewellen, 2000b), that had been selected for combined resistance to rhizomania, virus yellows, and agronomic traits. The sugarbeet germplasm was largely from C37 (PI590715) (Lewellen et al., 1985b), C78 (PI593671) (Lewellen et al., 1985a), C80 (PI593672) (Lewellen, 1997), and C82 (PI593675) (Lewellen, 1997). Resistance to rhizomania is conditioned by both *Rz* and factor(s) from C51 (*Bvm*) that gives a high level of resistance under high temperature conditions. During its development C67/2 has been tested as Y967 and Y167.

C69/2 was released previously as C69 (PI599341) in 1998 (Lewellen, 2000a). Since then, C69/2 has undergone two additional cycles of recurrent phenotypic selection. In both cycles, emphasis was placed on selecting mother roots for sucrose concentration, size, and conformation from field plants grown under rhizomania conditions, inoculated with virus yellows and sugarbeet *Erwinia*, and naturally infected with powdery mildew. C69/2 is predominantly the germplasm of C31/6 (PI590799) (Lewellen et al., 1978) with smaller amounts from C37, C46/2 (PI590800), C39 (PI583373) (Lewellen, 1995), C64 (McFarlane and Skoyen, 1965), and other sources. C69/2 is moderately resistant to virus yellows, bolting, powdery mildew, and *Erwinia*. It is moderately susceptible to curly top. During its development, C69/2 has been tested as breeding line numbers Y969 and Y169.

C78/3 was selected from C78/2 (PI593695) released in 1996 and C78 (PI593671) in 1994 (Lewellen, 1997). Since being released as C78/2, C78/3 has undergone three additional cycles of recurrent phenotypic selection. In each cycle, emphasis was on selecting mother roots for sucrose concentration, size, and conformation from field plants grown under rhizomania conditions, inoculated with virus yellows and sugarbeet *Erwinia*, and naturally infected with powdery mildew. C78/3 is predominantly the germplasm from curly top resistant breeding line C46/2 (PI590800) (Lewellen et al., 1985a). C78/3 is moderately resistant to virus yellows, bolting, powdery mildew, *Erwinia*, and *Beet curly top virus*. During its development, C78/3 has been tested as breeding line numbers R578, R578/2, R578%, R778, R778%, R978 and R178. Although handled as if completely self-sterile ($S^s S^s$), recent use of C78/3 progenitors as a recurrent parent in backcrossing programs has shown that some plants express various degrees of self-fertility.

C80/2 was selected from C80 (PI593672) (Lewellen, 1997), C80NB (PI593673), and C80-45 (PI593674) released in 1994. These sublines were recombined to produce C80/2. C80/2 has undergone four additional cycles of recurrent phenotypic selection. The first of these four cycles was for resistance to rhizomania in 4-month old plants within C80, C80NB, and C80-45. Selected plants from these lines were recombined into one population. In each of the next three cycles, emphasis was placed on selecting mother roots for sucrose concentration, size, and conformation from field plants grown under rhizomania conditions, inoculated with virus yellows and sugarbeet *Erwinia*, and naturally infected with powdery mildew. C80/2 was developed from a broad base of breeding lines in the virus yellows and multiple disease resistance program at Salinas. During its development, C80/2 has been tested as breeding line numbers R580, R580-45, R580NB, R780/2, R780-45, R980, and R180.

Lines C67/2, C69/2, C78/3, and C80/2 may be useful for continued line improvement and as sources of multiple disease resistant germplasm. These four lines represent a broad germplasm base and encompass much of the germplasm developed in the long term breeding program at Salinas. They account for much of the germplasm from the virus yellows (BYV/BWYV) breeding program that has been ongoing since 1955. On the basis of previous successes and evidence from progeny family evaluations (both S_1 and full sib), these lines may continue to be useful as sources from which to extract parental lines. U.S. Plant Variety protection will not be sought for these lines.

Lewellen, R.T. 2004. Registration of sugarbeet germplasm lines C927-4, C929-62, C930-19, and C930-35 with resistance to rhizomania, virus yellows, and bolting. Crop Sci. 44:359-361.

Sugarbeet (*Beta vulgaris* L.) germplasm lines C927-4 (Reg. no. GP-233, PI628756), C929-62 (Reg. no. GP-234, PI628757), C930-19 (Reg. no. GP-235, PI628758), and C930-35 (Reg. no. GP-236, PI628759) were developed by the USDA-ARS in cooperation with the Beet Sugar Development Foundation and the California Beet Growers Association. These lines were released in 2002. They are narrowly based each having been increased from one S_1 progeny (one selfed S_0 plant). They are multigerm (*MM*), self-fertile (S^f) diploids that segregate for genetic male sterility (*aa*) and resistance to rhizomania conditioned by *Rz1*. They have shown good general combining ability for sugar yield in experimental hybrids. In general, they show nonbolting tendency in over-wintered plantings and have tolerance to virus yellows (VY), caused by *Beet yellows virus* (BYV), *Beet western yellows virus* (BWYV), and *Beet chlorosis virus* (BChV). Except for the intermediate reaction for C930-35, all show high resistance to sugarbeet *Erwinia*, caused by *Erwinia carotovora* subsp. *betavasculorum* Thomson et al.

Lines C927-4, C929-62, C930-19, and C930-35 were identified and selected from a program designed to combine multiple disease resistance and factors for productivity. S_1 progeny evaluations followed by testcross hybrid evaluations were used. S_1 progeny evaluation is a useful plant breeding method for identifying and improving traits with additive genetic variance, e.g., most disease resistances and sucrose concentration. Breeding lines with self-incompatibility ($S^s S^s$) comprise most of the advanced, highly productive sugarbeet germplasm, however, they do not easily lend themselves to this breeding procedure. The program from which these lines were selected was designed to determine if self-incompatible lines could be worked quickly into an S_1

testing program. To accomplish this, self-incompatible lines were crossed onto genetic-male-sterile plants from self-fertile, genetic-male-sterile facilitated, random-mated populations that had been undergoing population improvement. These F_1 population or line hybrids were then used as the source of the S_0 plants to produce S_1 progenies. Because seed of population hybrids can be easily produced in large quantities, the S_0 plants can be selected after rigorous evaluation for one or more moderate to highly heritable traits. In this scheme, most of the S_0 plants will be pollen fertile (Aa) and their S_1 progenies will segregate $3A\_:1aa$, giving ample opportunity and flexibility for selecting materials to be used in a continuing line or population improvement program. With the exception of C930-19, only 6 yr were needed to go from the initial crosses to early generation lines with potential for development into parental lines for C927-4, C929-62, and C930-35.

C927-4 segregates for hypocotyl color (R). In addition to resistance to rhizomania conditioned by $Rz1$, resistance is also provided from factor(s) from *B. vulgaris* subsp. *maritima* (*Bvm*). C927-4 produces hybrids with intermediate sucrose concentration and high sugar yield. Relative performance of these hybrids is best when grown under rhizomania conditions. C927-4 is moderately susceptible to powdery mildew (caused by *Erysiphe polygoni* DC.) and *Beet curly top virus* (BCTV).

C927-4 was derived from a population cross between populations C918 (PI578079) (USDA, 1993) and 921. C918 is a multigerm, self-fertile, genetic-male-sterile facilitated, random-mated population. Self-fertile population 921 was developed from crosses between C918 and self-sterile lines R322Y3 and R322R4. Lines R322Y3 and R322R4 are similar to C51 (PI593694) (improved C50, PI538251) (Lewellen, 2000) that was developed from composite crosses between sugarbeet and *Bvm*. Theoretically, about 12% of C927-4 would be from *Bvm*. Population C918 is a source for the *Rz1* allele for resistance to rhizomania. C51 contributed additional factors that condition improved resistance and survivability of plants under the combined effects of severe rhizomania and high temperature stress. C927-4 possesses this type of resistance to rhizomania. From the F_1 population hybrid between genetic-male-sterile plants from C918 and fertile plants from 921, individual S_0 plants were selected for sucrose concentration under VY inoculated (BYV/BWYV/BChV) conditions and selfed under bags to produce S_1 progeny families. These S_1 progenies were evaluated for resistance to rhizomania at Salinas and Brawley, CA, for performance under VY inoculated conditions at Salinas and Davis, CA and for bolting tendency at Salinas. On the basis of these tests, S_1 progenies were selected, increased in isolation, and testcrossed to a monogerm, cytoplasmic male-sterile line. Line 9927-4VY was selected based on the performance of its experimental hybrid and increased through its genetic-male-sterile segregants to produce line 1927-4 that was released as C927-4.

C929-62 has red hypocotyls (RR) and near seed maturity has reddish stems and seedballs. It has moderately high resistance to powdery mildew and is moderately susceptible to BCTV and downy mildew caused by *Peronospora farinose* (Fr:Fr.) Fr.]. C929-62 produces hybrids with intermediate sugar concentration and high sugar yield.

C929-62 was derived from a population cross between genetic-male-sterile plants from population C918 and C76-89-18 (PI593699). Self-sterile line C76-89-18 was advanced from one full-sib progeny that was susceptible to rhizomania but had high sugar yield combining ability and resistance to VY, *Erwinia*, and bolting. It was selected from C31/6 (PI590799) type germplasm.

From the F_1 population hybrid, individual S_0 plants were selected for sucrose concentration under VY inoculated conditions and were selfed under bags to produce S_1 progenies. These S_1 progenies were evaluated at Salinas and Davis for performance under virus yellows inoculated conditions and at Salinas for components of sugar yield, resistance to rhizomania, and nonbolting tendency. On the basis of these tests, S_1 progenies were selected, increased, and testcrossed to a monogerm, CMS tester. Line 9929-62VY was selected for further evaluation based on the performance of its experimental hybrid. Line 9929-62VY was increased through its male-sterile segregants to produce line 1929-62 that was released as C929-62.

C930-19 segregates for hypocotyl color. It is moderately resistant to BCTV and powdery mildew and has very high nonbolting tendency. In tests at Salinas and Brawley, its hybrids have moderate to high sugar concentration and sugar yield.

C930-19 was derived from a population cross made in 1995 between population C918 and breeding line C78 (PI593671). C78 is a rhizomania resistant version of C46/2 (PI590800). Self-sterile C46/2 has moderate BCTV resistance and has been an important source of pollinators used commercially in California. From the F_1 population hybrid, individual S_0 plants were selected for resistance to rhizomania and were selfed to produce S_1 progenies. These S_1 progenies were evaluated at Salinas for components of sugar yield and for resistance to bolting, rhizomania, powdery mildew, and VY. On the basis of these tests, S_1 progenies were selected, increased, and testcrossed to a monogerm, CMS tester. Line 8930-19 was chosen from among this group for further evaluation based on the performance of its experimental hybrid. Over-wintered stecklings from Oregon of 8930-19 were transplanted into a field isolation plot at Salinas. In the absence of an artificially extended photoperiod, stecklings of 8930-19 were very slow to bolt and some plants did not flower. During seed harvest, 30 of these non-flowering plants were saved out of an initial 210 stecklings, regrown in the greenhouse, and vernalized for 140 d, then replanted into a greenhouse isolation chamber with a 24-h photoperiod. Under these conditions, this nonbolting selection from line 8930-19 produced seed. This seed was harvested in bulk without regard to male sterile segregants and called 1930-19. Line 1930-19 was reselected for resistance to rhizomania and selected plants were increased through its genetic-male-sterile segregants to produce 2930-19. Line 2930-19 was released as C930-19.

C930-35 has green hypocotyls (*rr*), is moderately resistant to BCTV and powdery mildew and has high sucrose concentration. C930-35 produces hybrids with high sugar concentration but moderate root and sugar yields.

C930-35 was derived from a population cross made in 1996 between genetic-male-sterile plants from one component of population CZ25 (PI599343) and breeding line C78. This component of CZ25 was a multigerm, self-fertile, genetic-male-sterile facilitated, random-mated population. It was developed from crosses between breeding sources similar to C918 and high sucrose accessions from Poland. About 25% of the germplasm of C930-35 would be Polish. The Polish germplasm was from $2n = 2x = 18$ chromosome, multigerm, self-incompatible (S^sS^s), type-ZZ lines accessed from Dr. A. Szreder, Hodowla Buraka Cukrowego, Poland, in 1988 for use in the Salinas breeding program. A composite of nine Polish accessions were crossed to genetic-male-sterile plants from a progenitor of population C918 to ultimately produce population CZ25. From the F_1 population hybrid between CZ25 and C78, individual S_0 plants were selected for resistance to rhizomania and

were selfed in bags to produce S_1 progenies. These S_1 progenies were evaluated at Salinas for components of sugar yield and resistance to bolting, rhizomania, and powdery mildew. At both Salinas and Davis, they were evaluated for sugar yield under virus yellows inoculated conditions. On the basis of these tests, S_1 progenies were selected, increased, and testcrossed to a monogerm, CMS tester. Line 9930-35 was selected for further evaluation based on the performance of its experimental hybrid. Line 9930-35 was increased through its male-sterile segregants to produce line 1930-35 that was released as C930-35.

Lines C927-4, C929-62, C930-19, and C930-35 may be useful as germplasm sources for further improvements and as sources of combined disease and bolting resistance in highly productive backgrounds. They need to be evaluated as early generation lines for the potential development of pollinators for commercial hybrids. U.S. Plant Variety Protection will not be sought for these lines.

Lewellen, R.T. 2004. Registration of CP03, CP04, CP05, and CP06 sugarbeet germplasm with resistance to powdery mildew, rhizomania, and other diseases. Crop Sci. 44:1886-1887.

Sugarbeet (*Beta vulgaris* L.) germplasm lines CP03 (Reg. no. GP-240, PI632284), CP04 (Reg. no. GP-241, PI632285), CP05 (Reg. no. GP-242, PI632286), and CP06 (Reg. no. GP-243, PI632287) were developed by the USDA-ARS in cooperation with the Beet Sugar Development Foundation (BSDF) and the California Beet Growers Association. They were released in 2003.

CP03 and CP04 are multigerm (*MM*), self-sterile (*S^sS^s*), germplasm lines that segregate for resistance to powdery mildew (*Pm*) (Lewellen & Schrandt, 2001; Janssen et al., 2003) caused by *Erysiphe polygoni* DC. (syn. *E. betae* Weltzien) and rhizomania (*Rz1*) (Lewellen et al., 1987) caused by *Beet necrotic yellow vein virus* (BNYVV). CP03 and CP04 have identical developmental histories except for the *B. vulgaris* subsp. *maritima* source of resistance to powdery mildew. Resistance within CP03 is from WB97 (PI546394) and CP04 is from WB242 (PI546413). Through the BC_3F_1 generation, CP03 was the same as CP01 (PI610490) and CP04 as CP02 (PI610491) (Lewellen, 2000). For the fourth backcross, C78/3 (PI628752) (Lewellen, 2004) was used. For backcrosses five and six, C37 (PI590715) (Lewellen et al., 1985) was again used as the recurrent parent. Theoretically, CP03 and CP04 would have approximately 87% of their germplasm from C37, 12% from C78/3, and 1% from the wild beet source of resistance to powdery mildew. Starting from the BC_4F_1 generations, in general, individual plants were selected from the backcross families for resistance to powdery mildew and rhizomania and pair-crossed in the greenhouse to the recurrent parent. For the BC_6F_1 families, individual pair-crosses were evaluated in the field at Salinas in a March planting under natural powdery mildew and rhizomania infected conditions. Individual plants from within these families were selected in November for high resistance to both powdery mildew and rhizomania and for nonbolting. Within sets of families from each source of resistance, selected plants were combined and increased in mass to produce BC_6F_2 populations released as CP03 and CP04. CP03 is from seed lot P227 and had been developed and tested as lines P327, P127, P027, P917, and P815. CP04 is from seed lot P228 and had been developed and tested as lines P328, P128, P028, P918, and P816. Other than for powdery mildew and rhizomania, disease resistance and agronomic traits of CP03 and CP04 should be similar to C37, but, in the BC_6F_1 families, obvious, visual differences were evident. Segregation for annualism and *B. vulgaris* subsp. *maritima* coloring patterns still occurred. In addition, in tests in Brawley, CA, CP04 was more

resistant to rhizomania under high temperature conditions than CP03 or C78/3 and appeared to be tolerant to phytotoxemia from the feeding of leafhoppers [*Empoasca fabae* (Harris) and *E. solana* DeLong] retaining its canopy longer in a full, dark green condition. In preliminary tests, CP04 appeared to segregate for partial resistance to sugarbeet cyst nematode (*Heterodera schachtii* Schmidt). In the BSDF nursery near Kimberly, ID, CP03 and CP04 were slightly more susceptible to *Beet curly top virus* (BCTV) than C37. In tests at Salinas in 2003, they had similar reactions to *Beet chlorosis virus* (BChV) and *Erwinia carotovora betavasculorum* Thomsen et al. and for bolting tendency. CP03 and CP04 should be useful as enhanced sources of resistance to powdery mildew found in *B. vulgaris* subsp. *maritima* and for genetic and plant pathological research.

CP05 and CP06 are multigerm (*MM*), self-sterile (S^sS^s) germplasm lines that segregate for resistance to powdery mildew (*Pm*) and rhizomania (*Rz1*). CP05 and CP06 have identical developmental histories except for the *B. vulgaris* subsp. *maritima* source of resistance to powdery mildew. Resistance within CP05 is from WB97 and CP06 is from WB242. Through the BC_3F_2 generation, CP05 was the same as CP01 (PI610490) and CP06 as CP02 (PI610491). From backcross four through seven, the recurrent parent for CP05 and CP06 was C78/3. Usually, the lines were advanced from seed produced on C78/3 or from reciprocal pairs that had identical appearance in field plots. Starting from the BC_3 generations, in general, individual plants were selected from the backcross families for resistance to powdery mildew and rhizomania and pair-crossed under paper bags in the greenhouse to C78/3. For the BC_7F_1 families, individual pair-crosses were evaluated in the field at Salinas in a March planting under natural powdery mildew and rhizomania infected conditions. Individual plants from within these families were selected in November for high resistance to powdery mildew, resistance to rhizomania, and for nonbolting. Within sets of families from each source of resistance, selected plants were combined and increased in mass to produce the BC_7F_2 populations released as CP05 and CP06. CP05 is from seed lot P229 and had been developed and tested as lines P329, P129, P029, P919, and P809. CP06 is from seed lot P230 and had been developed and tested as lines P330, P130, P030, P920, and P810. In addition to powdery mildew resistance conditioned by *Pm*, CP05 and CP06 likely have the moderate slow-mildewing type of resistance derived from C78/3 in contrast to CP03 and CP04. The other disease resistance and agronomic traits of CP05 and CP06 should be similar to C78/3 but obvious visual differences remain. Up through BC_7F_1 families, annualism and coloring patterns of *B. vulgaris* subsp. *maritima* lines WB97 and WB242 still occurred. CP05 and CP06 appeared to be moderately resistant to BCTV but slightly more susceptible than C78/3. They had similar reactions as C78/3 to BChV and sugarbeet *Erwinia*. CP05 and CP06 should be useful as enhanced sources of resistance to powdery mildew originally found in *B. vulgaris* subsp. *maritima* and for genetic and plant pathological research.

Lewellen, R.T. 2004. Registration of CP07 and CP08 sugarbeet germplasms with resistance to powdery mildew, rhizomania, and other diseases. Crop Sci. 44:2276-2277.

Sugarbeet (*Beta vulgaris* L.) germplasm lines CP07 (Reg. no. GP-244, PI632288) and CP08 (Reg. no. GP-245, PI632289) were developed by the USDA-ARS in cooperation with the Beet Sugar Development Foundation (BSDF) and the California Beet Growers Association. They were released in 2003. CP07 and CP08 are multigerm (*MM*), germplasm lines that segregate for resistance to powdery mildew (*Pm*) (Lewellen & Schrandt, 2001) caused by *Erysiphe polygoni* DC. and

rhizomania (*Rz1*) (Lewellen et al., 1987) caused by *Beet necrotic yellow vein virus*. In addition CP07 and CP08 may have resistance to rhizomania conditioned by factors from *B. vulgaris* subsp. *maritima* (Lewellen & Whitney, 1993). They segregate for hypocotyl color and are likely self-sterile (S^sS^s) although segregation for self-fertility (S^f) is possible.

As a line and in experimental hybrids CP07 shows moderate resistance to sugarbeet *Erwinia* caused by *E. carotovora betavasculorum* Thomsen et al. and bolting tendency. It is intermediate for reaction to *Beet curly top virus*, similar to C78/3 (Lewellen, 2004a). It may show tolerance or have reduced infestation counts to sugar beet cyst nematode (*Heterodera schachtii* Schmidt) based upon field observations (Lewellen, unpublished). At Salinas, in the absence of rhizomania, it has lower sugar yield and sucrose concentration than the mean of four commercial hybrid checks. Under rhizomania, it has higher sugar yield and equal sucrose concentration than the mean of these same rhizomania resistant checks. At Brawley, CA, under both rhizomania and nonrhizomania conditions, it had higher sugar yield and sucrose concentration than these rhizomania resistant commercial checks and other experimental lines and hybrids that depended solely upon the *Rz1* factor for resistance to rhizomania. At Brawley, the late season survival and appearance score was very good and similar to C927-4 (PI628756) (Lewellen, 2004b). In the bolted phase, CP07 segregates for determinate growth of stems. As far as is known, this is a previously undescribed morphological trait that causes the stem to abruptly end in a flower or cluster of flowers. On the bolted stems, most leaf axils have only flowers but not lateral branches. From one to many internodes are formed before stem termination.

At Salinas, in 1999, 15 individual plants were selected from among three backcross families,. These mother roots were selected for resistance to rhizomania, high resistance to powdery mildew, and nonbolting. Earlier in 1999, these same three backcross families were observed to segregate for high resistance and survival to rhizomania under high temperature, severe rhizomania conditions in Imperial Valley tests. The recurrent parents leading to CP07 were C37 (PI590715) (Lewellen et al., 1985), C72 (PI599342), and C78/3 (PI628752). The final two backcrosses were to C78/3. The donor parents had germplasm from *B. vulgaris* subsp. *maritima* that contributed resistance to powdery mildew and rhizomania. It is estimated that CP07 has about 72% of its germplasm from C78/3, 24% from C37, 3% from *B. vulgaris* subsp. *maritima* through C72 from C51 (PI593694) (Lewellen, 2000b), and 1% from both WB97 (PI546394) and WB242 (PI546413) (Lewellen, 2000a). Of the six parental plants in the final backcross, three plants were C78/3, and three had C51 germplasm in their background. Of the latter three plants, two also had germplasm from WB97 and one from WB242. It is believed that resistance to powdery mildew (*Pm*) was derived from WB97 and/or WB242 (Lewellen & Schrandt, 2001) and resistance to rhizomania from C78/3 (*Rz1*) and C51 and/or WB97 and WB242 for high resistance and survival under high temperature, severe rhizomania conditions. The 15 plants selected in 1999 were increased in mass in 2000 to produce P007/8. Line P007/8 was reselected in 2001 under natural powdery mildew, rhizomania, and cyst nematode infested conditions for resistance to powdery mildew and rhizomania and freedom from infestation with nematodes to produce P207/8. Line P207/8 was released as CP07.

CP08 shows intermediate nonbolting tendency. At Brawley, CA, under rhizomania conditions, it has higher sugar yield and sucrose concentration than lines with similar germplasm and parentage. At Brawley, the late season survival score is superior to most other entries. Under moderate to severe rhizomania and unknown soil-borne problems at Brawley, the canopy of CP08 remains dark

green. This appears to be due to a combination of high resistance to rhizomania and/or other soil-borne factors, e.g., possibly sugar beet cyst nematode, high resistance to powdery mildew, and resistance to phytotoxemia from the feeding of *Empoasca* leafhoppers (*E. fabae* Harris and *E. solana* DeLong).

CP08 was increased from one full-sib line that in progeny tests in 2000 at Brawley and Salinas, segregated for high resistance to powdery mildew, resistance to rhizomania, and under severe rhizomania, segregated for very good appearance and survival scores under high temperatures. This full-sib progeny resulted from backcrosses to transfer and combine *Pm* and *Rz1*. A number of powdery mildew resistant plants from CP02 (Lewellen, 2000a) were backcrossed to plants of C78/3, the source of *Rz1*. Individual plants from this series of backcrosses that appeared to be resistant to powdery mildew and rhizomania were backcrossed by paired crosses in the greenhouse under paper bags to plants from C37. Backcross P918-6 was selected from progeny tests in 2000 and increased to produce line P118-6. Seed of P118-6 was released as CP08 and further tested as P318-6. About 2% of CP08 was derived from WB242, 25% from C78/3, and 73% from C37. Under severe rhizomania and high temperature conditions, CP08 is strikingly different from C78/3 and C37 for resistance to rhizomania, powdery mildew, and the feeding effects of *Empoasca*. Under these conditions at Brawley, CP08 has a very desirable, dark green appearance that gives the canopy a "stay-green" tendency.

Lines CP07 and CP08 should be evaluated as sources from which to develop potential pollinators for high performing, disease and bolting resistant hybrids. These lines may be useful as a combined source of high resistance to powdery mildew and rhizomania. They need to be evaluated further as a potential source of tolerance to cyst nematode and *Empoasca* leafhoppers.

Liu, H.-Y. and J.J. Gallian. Identification of a new strain of the rhizomania virus and its potential impact on the sugar beet industry. Procedings of the winter commodity schools-2004, University of Idaho, Cooperative Extension System, Volume 36:233-235. 2004.

Rhizomania is one of the most destructive diseases of sugar beet, not only because it causes a severe loss in root yield and sugar content but also it is difficult to control. Rhizomania is widely distributed in most sugar beet-growing areas world wide. This disease is caused by *Beet necrotic yellow vein virus* (BNYVV) and vectored by the plasmodiophorid fungus *Polymyxa betae*. The presence of rhizomania could result in a total loss for a sugar beet crop. At the present time, the most effective control measure is to use partially resistant sugar beet cultivars based upon single dominant genes against this devastating disease. In the summer of 2002 and 2003, several sugar beet fields with BNYVV-resistant cultivars in the Imperial Valley, California were observed with severe rhizomania symptoms. Standard soil baiting with sugar beet seedlings followed by enzyme-linked immunosorbent assay (ELISA) were conducted. Resistant varieties grown in regular BNYVV-infested soil remained resistant. In contrast, when grown in Imperial Valley BNYVV-infested soil all resistant varieties tested susceptible according to elevated ELISA values. From the soil testing, results suggested that resistance had been compromised in Imperial Valley.

Three pathotypes of BNYVV have been reported. Pathotype A has world wide distribution. It has been found in most sugar beet growing countries including the United States. Pathotype B was

observed only in Germany and the upper Rhine Valley in France. Pathotype A and pathotype B are very similar and the virus possesses four RNAs. Pathotype P found in the region around the French town of Pithiviers and East Anglia in the United Kingdom contain a fifth RNA. Experimental evidence has shown that this strain seems to be more aggressive than the A and B pathotypes and can infect partially resistant beet varieties.

We collected BNYVV-infested soil from Imperial Valley. After soil baiting tests we mechanical inoculated the infected roots to a local lesion host *Chenopodium quinoa* plants. From each single local lesion isolate we did host range tests. Based on the host reaction we have isolated eight distinct BNYVV isolates from Imperial Valley (IV-BNYVV). In order to find out whether IV-BNYVV isolates contain RNA-5, we used reverse transcription-polymerase chain reaction (RT-PCR) technique. Our results indicate that the BNYVV isolates from Imperial Valley did not contain RNA-5.

To confirm that Imperial Valley does not have P-pathotype of BNYVV, we did single-strand conformation polymorphism analysis (SSCP). From our experiment, results indicate that IV-BNYVV isolates banding patterns from RNA 1 and RNA 2 were identical to A-pathotype and different from P-pathotype.

IV-BNYVV isolates do not contain RNA-5 as determined by RT-PCR. In SSCP analyses, all of the IV-BNYVV isolates the banding patterns were identical to A-pathotype. We concluded that the resistance-breaking BNYVV isolates from Imperial Valley likely had evolved from the original existing A-pathotype.

Possible Impact of the New Strain on the Idaho Sugar beet Industry

Rhizomania was first diagnosed in the Imperial Valley of California in 1990, and the new strain was found 12 years later in 2002. In Idaho, the disease was first found in the Magic Valley near Rupert in 1992 on 670 acres. In 1996, it was identified in the Treasure Valley near Middleton. Since 1996 Idaho growers have been planting rhizomania resistant varieties in known infested fields. We estimate that approximately 89,000 acres (40%) in the Amalgamated Sugar Company growing area are now infested. This year it has been 12 years in the Magic Valley and 8 years in the Treasure Valley since the disease has been first diagnosed.

Short rotations increase selection pressure on the pathogen and reduce the time before vew strains become dominant in the population. In Idaho, far too many growers are continuing to plant sugar abets on one- and two-year rotations. In rhizomania-infested fields a three-year rotation is standard and four years in preferred. With four-year rotation, it will obviously take four times as long for the process to occur. Based on UI studies in Rupert, Idaho, from 1995 through 1999, lengthening the rotation from one year to four can increase yield under severe rhizomania conditions from 16.1 to 30.7 tons/acre.

The current situation is considerably different than it was in 1992. At that time, resistance to BNYVV had been discovered nine years before, and rhizomania-resistant varieties were already being planted in California. At this time, there has not yet been resistance identified to the new pathogenic strain of BNYVV. Lengthening the sugar beet rotation is the best way to gain time for

the identification of resistance, breeding new resistant varieties and developing other management practices.

Liu, H. Y., J.L. Sears, and R.T. Lewellen. Emergence of resistance-breaking isolates of Beet necrotic yellow vein virus in the Imperial Valley, California. Phytopathology 94:S62. 2004.

Beet necrotic yellow vein virus (BNYVV) is the causal agent of rhizomania disease of sugar beet. The virus is transmitted by the soil-borne fungus *Polymyxa betae*. The disease can only be controlled by the use of resistant cultivars. During 2002 and 2003 in the Imperial Valley of California partially resistant sugar beet cultivars with *Rz1* allele developed against this devastating disease seem to be compromised. Distinct BNYVV isolates were isolated from infected sugar beet roots (IV-BNYVV) by single local lesion isolation. These isolates do not contain RNA-5 as determined by RT-PCR. From the banding patterns of single-strand conformation polymorphism analyses we concluded that the resistance-breaking BNYVV isolates from Imperial Valley had likely evolved from the original existing A-type. The pathogenicity of IV-BNYVV isolates was studied. PCR products from coat protein (RNA-2) and 25-kDa protein (encoded by BNYVV-RNA-3, involved in symptom expression) of IV-BNYVV isolates were sequenced. Sequence alignments revealed only minor amino acid changes compared to the existing A-type of California BNYVV isolates.

Liu, H. Y., J.L. Sears, and R.T. Lewellen. P-pathotype of rhizomania in sugar beet has not been identified in the Imperial Valley, California. The California Sugar Beet p.12-13, 25. 2004.

Rhizomania is one of the most economically important diseases of sugar beet. This disease is caused by *Beet necrotic yellow vein virus* (BNYVV) and vectored by the soil-borne fungus *Polymyxa betae*. Partially resistant sugar beet cultivars based upon single dominant genes have been developed against this devastating disease. During 2002 and 2003 in the Imperial Valley of California sugar beet fields with a BNYVV-resistant cultivar were observed with severe rhizomania symptoms, suggesting that resistance had been compromised. Standard soil baiting with sugar beet plants followed by ELISA tests were used to diagnose virus occurrence and reaction. Resistant varieties grown in regular BNYVV-infested soil remained resistant. In contrast, when grown in Imperial Valley BNYVV-infested soil all resistant varieties tested susceptible according to elevated ELISA values. Eight different BNYVV isolates have been isolated from Imperial Valley soil (IV-BNYVV) by single local lesion isolation. IV-BNYVV isolates did not contain RNA-5 as determined by RT-PCR. In single-strand conformation polymorphism analyses all the isolates the banding patterns were identical to A-type and different from P-type. From our results indicate the resistance-breaking BNYVV isolates derived from existing A-type.

McGrath, J.M. and R.T. Lewellen. 2004. Registration of EL0204 sugarbeet germplasm with smooth-root and resistance to rhizomania. Crop Sci. 44:1032-1033.

See report by McGrath.

Segev, L., W.M. Wintermantel, J.E. Polston, and M. Lapidot. First Report of *Tomato chlorosis virus* in Israel. *Plant Disease* 88:1160.

In December 2003, symptoms were observed in greenhouse tomato plants in Bet Dagan, Israel, which resembled those of *Tomato chlorosis virus* (ToCV), a crinivirus common in the southeastern United States and Southern Europe (2, 3). Middle-aged leaves showed interveinal chlorosis while more mature leaves showed more intense interveinal chlorosis with some interveinal bronzing. Symptoms were associated with the presence of *Bemisia tabaci*, an efficient vector of ToCV. Total nucleic acids were extracted (1) from middle-aged and mature leaves from two symptomatic plants, as well as from healthy tomato, *Physalis wrightii* infected with ToCV, and *Nicotiana benthamiana* infected with *Tomato infectious chlorosis virus* (TICV), another crinivirus that produces identical symptoms on tomato. Extracts were tested by hybridization with probes specific to the coat protein gene of ToCV and to the HSP70h gene of TICV. Hybridization results identified the presence of ToCV in all samples from symptomatic tomato plants and ToCV-infected *P. wrightii*, but not in those from healthy tomato or TICV-infected *N. benthamiana*. TICV was only detected in TICV-infected *N. benthamiana*. Extracts were also subjected to RT-PCR using primers specific to the coat protein gene of ToCV (NCBI No. AY444872; Forward primer: 5' ATGGAGAACAGTGCCGTTGC 3'; Reverse Primer: 5' TTAGCAACCAGTTATCGATGC 3'). All samples from symptomatic tomato and ToCV-infected *P. wrightii* produced bands of the expected size, but no bands were produced from extracts of healthy tomato. Laboratory results and observed symptoms confirm the presence of ToCV in symptomatic tomatoes. To our knowledge, this is the first report of ToCV in Israel.

Tsai, W. S., S.L. Shih, S.K. Green, P. Hanson, and H.Y. Liu. 2004. First report of the occurrence of *Tomato chlorosis virus* and *Tomato infectious chlorosis virus* in Taiwan. Plant Disease 88:311.

Pronounced yellowing symptoms on the lower leaves of tomato plants, similar to those caused by nitrogen deficiency, were observed in the spring of 1998 in the Asian Vegetable Research and Development Center and in farmers' fields in southern Taiwan. However, the brittleness of the discolored leaves, occasional upward leaf rolling, and abundance of whiteflies on these plants suggested the involvement of *Tomato chlorosis virus* (ToCV) and *Tomato infectious chlorosis virus* (TICV) that belong to the group of whitefly-transmitted, phloem-limited criniviruses (Family *Closteroviridae*). Leaves of symptomatic and healthy plants were collected, and total nucleic acids were extracted from 0.2 g of leaf tissue. The total nucleic acids were precipitated by ethanol and dissolved in 160 µl of sterile water. Eight microliters of total nucleic acids were absorbed on positively charged nylon membranes (Roche Diagnostic GmbH, Roch Applied Science, Germany). Two digoxigenin-labeled riboprobes, transcribed from pTIC8-44 (complementary to the 3'-end region of TICV RNA 1) and pToCV 78 (corresponding to the coat protein region of ToCV RNA 2), were used in hybridization tests to detect TICV and ToCV respectively. Six of seventeen symptomatic tomato plant samples were positive with the ToCV probe, whereas none of the 13 samples reacted with the TICV probe. Similar symptoms as described above for tomato were observed on zinnia plants in the same locations. Five of eight zinnia samples gave a positive reaction with the ToCV probe. One of the ToCV positive samples also gave a positive reaction with the TICV probe. Electron microscopic examination from leaf-dip preparations of ToCV-positive

leaf tissues, stained in 1% uranyl acetate, showed the presence of flexuous filamentous particles approximately 800 to 850 nm long. To our knowledge, this is the first evidence of the presence of ToCV and TICV in zinnia and ToCV in tomato in Taiwan.

Tzanetakis, I.E., A.B. Halgren, W.M. Wintermantel, K.E. Keller, and R.R. Martin. 2004. Two criniviruses are associated with the strawberry pallidosis disease. *Acta Horticulturae* **(online) < http://www.actahort.org/books/656/656_1.htm>**

Pallidosis, a disease attributed to a graft-transmissible agent is the focal point of this study. Two viruses belonging to the *Closteroviridae* family, genus *Crinivirus* that can cause pallidosis symptoms on strawberry indicator plants have been identified. A previously unidentified virus designated as *Strawberry pallidosis associated virus* (SPaV) is the predominant virus in pallidosis positive plants, while the second virus is *Beet pseudo-yellows virus* (BPYV). The genomes of both viruses have been sequenced fully and phylogenetic analyses indicate that the two viruses are more closely related than any other crinivirus found in the database. Epidemiological studies have demonstrated that the greenhouse whitefly, *Trialeuroides vaporariorum,* is an efficient vector of SPaV. Protocols for molecular detection of both viruses using reverse transcription-polymerase chain reaction (RT-PCR) have been developed. The recombinant major coat protein of SPaV has been expressed in bacteria and polyclonal antibodies to the virus have been developed that facilitate detection of the virus in tissue blot immunoassays (TBIA). The potential of seed and pollen transmission of SPaV was examined as an alternative mode of transmission of the virus. The geographical distribution of both viruses in the major strawberry producing regions of the United States has been examined.

Wintermantel, W.M. 2005. Co-infection of *Beet mosaic virus* with beet yellowing viruses leads to increased symptom expression on sugarbeet. *Plant Disease* **89: 325-331.**

Three distinct aphid-transmitted viruses associated with a yellowing disease on sugar beet were examined in single and mixed infections for the effects of virus interactions on plant weight, rate of symptom development and virus concentration. Sugarbeet lines exhibiting different degrees of susceptibility to the virus yellows complex were inoculated with either one, two or all three viruses. Severe stunting, as measured by fresh plant biomass, was observed during mixed infections with *Beet yellows virus* (BYV) and *Beet mosaic virus* (BtMV), compared to single infections of these viruses. In addition, the overall rate of appearance of *Beet western yellows virus* (BWYV) symptoms increased during co-infection with BtMV. Synergistic effects on stunting severity, as measured by plant biomass, were more pronounced in susceptible beet lines, but similar patterns also were observed in lines exhibiting tolerance to virus yellows. Relative concentrations of viruses were compared among single and mixed infections using dot blot hybridization with virus specific probes, and quantified by phosphorimage analysis. Titers of all three viruses increased as a result of co-infection compared with single infections.

Wintermantel, W.M., G.C. Wisler, A.V. Karasev, and H.-Y. Liu. Genome organization and sequence of *Tomato chlorosis virus*. Phytopathology 94 (6S): S111.

The genome of *Tomato chlorosis virus* (ToCV) was cloned and the complete nucleotide sequence of the virus determined and compared with related crinivirus species. RNA1 is organized into 3 ORFs, and encodes genes involved in replication of viral RNA based on homology to other viral replication factors. RNA 2 is composed of 7 ORFs encoding a hsp70 homolog and two proteins involved in encapsidation of viral RNA, referred to as the coat protein and minor coat protein. Sequence homology between ToCV and other criniviruses varies throughout the viral genome. The ORF encoding the minor coat protein of ToCV, which forms part of the "rattlesnake tail" of virions and may be involved in determining the unique, broad vector transmissibility of ToCV, is larger than the comparable LIYV gene by 651 nucleotides. Among criniviruses sequenced, considerable variability exists in the size of some viral proteins. Analysis of these differences with respect to biological function may provide insights into the role crinivirus proteins play in virus infection and transmission.

Yu, M.H. and R.T. Lewellen. 2004. Registration of root-knot nematode-resistant sugarbeet germplasm M6-2. Crop Sci. 44:1502-1503.

Sugarbeet (*Beta* vulgaris L.) germplasm M6-2 (Reg. no. GP ..., PI632234) was developed by the USDA-ARS, Salinas, CA, in cooperation with the California Beet Growers Association, Ltd., Stockton, CA, and released in December 2002. M6-2 is highly resistant, if not immune, to root-knot nematode (*Meloidogyne* spp.)

M6-2 was produced by inter-pollinating more than 30 plans selected from the fifth backcross generation progeny of hybrids between M66 (PI 586688; Yu, 1996) and cultivated sugarbeet lines, including C37 (PI 590715; Lewellen et al., 1985) and C78 (PI 593671; Lewellen, 1997). F_1BC_5 plants with root-knot resistance were intercrossed. Nematode resistant F_2BC_5 plants were individually test crossed to a susceptible line. F_2 plants that performed well in test crosses and appeared to be homozygous for resistance were intercrossed to produce M6-2. M6-2 is a multigerm, biennial, self-incompatible germplasm that is heterogeneous for plant type and hypocotyl color. Approximately 25% of the seedlings have green hypocotyls. Root size and root conformation are not as uniform as the recurrent parents. Due to its wild beet ancestry, roots of M6-2 are often sprangled.

The M6-2 germplasm is resistant to multiple species of root-knot nematode, including *M. incognita* (Kofoid and White) Chitwood, *M.javanica* (Treub) Chitwood, *M. arenaria* (Neal) Chitwood, *M.hapla* Chitwood, *M.chitwoodi* Golden et al., and *M.fallax* Karssen, based on J2 larval inoculation studies in the greenhouse and monoxenic *M.incognita* and *M.javanica* infested field trials (Yu et al., 1999; Yu and Roberts, 2002). The level of resistance to root-knot nematode in M6-2 and M6-1 (PI 613165; Yu, 2001), a first generation backcross progeny of M66, appear to be similar. However, M6-1 is a self-compatible line with green hypocotyls, and taproots tend to be more sprangled than roots of M6-2.

BOOK CHAPTERS:

Wintermantel, W.M. 2004. Emergence of Greenhouse Whitefly (*Trialeurodes vaporariorum*) transmitted criniviruses as threats to vegetable and fruit production in North America. *APSnet feature* (online) http://www.apsnet.org/online/feature/whitefly/

Mutschler, M.A. and Wintermantel, W.M. 2005. Reducing Virus Associated Crop Loss Through Resistance To Insect Vectors. In: Natural Resistance Mechanisms to Viruses. Loebenstein, G. ed. Springer, New York. (in press).

POPULAR (non-peer reviewed) PUBLICATIONS:

Wintermantel, W.M. 2004. Understanding beet curly top: a complex disease that has persisted for over a century. *The California Sugarbeet-2003*. pp14-15, 25.

Wintermantel, W.M., Tzanetakis, I.E., Martin, R.R. 2004. Management of whitefly transmitted viruses in strawberry. *The Pink Sheet Strawberry News Bulletin.*

Wintermantel, W.M. and Grube, R.C. 2004. Lettuce Dieback: A growing virus problem in the West. *Vegetables West.* October issue, pp12-14.

STUDY OF NEW PATHOTYPES OF RHIZOMANIA IN THE UNITED STATES

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SUMMARY

Beet necrotic yellow vein virus (BNYVV) is the causal agent of rhizomania disease of sugar beet. The virus is transmitted by the soil-borne fungus *Polymyxa betae*. The disease can only be controlled by the use of resistant cultivars. During 2002 and 2003 in the Imperial Valley of California, partially resistant sugar beet cultivars with *Rz1* allele developed against this devastating disease seem to be compromised. Distinct BNYVV isolates were isolated from infected sugar beet roots (IV-BNYVV) by single local lesion isolation. These isolates do not contain RNA-5 as determined by RT-PCR. From the banding patterns of single-strand conformation polymorphism analyses we concluded that the resistance-breaking BNYVV isolates from Imperial Valley had likely evolved from the original existing A-type. The pathogenicity of IV-BNYVV isolates was studied. PCR products from coat protein (RNA-2) and 25-kDa protein (encoded by BNYVV-RNA-3, involved in symptom expression) of IV-BNYVV isolates were sequenced. Sequence alignments revealed only minor amino acid changes compared to the existing A-type of California BNYVV isolates.

INTRODUCTION

Rhizomania is one of the most economically important diseases of sugar beet and is widely distributed in most sugar beet growing areas worldwide. This disease is caused by *Beet necrotic yellow vein virus* (BNYVV) (Tamada and Baba, 1973; Tamada, 1975) and vectored by the plasmodiophorid *Polymyxa betae* Keskin (Fujusawa and Sugimoto, 1976). Most sugar beet production areas are dependent upon resistant sugar beet cultivars to control this devastating disease.

There are three major strain groups of BNYVV that have been reported (Kruse et al., 1994; Koenig et al., 1995; Koenig and Lennefors, 2000). Pathotype A was found in most countries. Pathotype B was observed in Germany and the upper Rhine Valley in France. Pathotype A and pathtype B contained four genomic RNAs. Pathotype P contained a fifth RNA seems to be more aggressive, and has so far been found in the region around the French town of Pithiviers and East Anglia in the UK which contained a fifth RNA. Other more infective strains of BNYVV have been found in Kazakhstan, China, and Japan. Experimental evidence from Europe, Japan, and the UK has shown that p-pathotype can infect partially resistant beet varieties. The different BNYVV pathtypes could be distinguished by means of Restriction fragment length polymorphism (RFLP) and single strand conformation polymorphism (SSCP) analysis of RT-PCR products (Kruse, et al., 1994; Koenig et al., 1995).

In the summer of 2002, three sugar beet fields planted with a BNYVV-resistant cultivar in the Imperial Valley of California were observed to exhibit severe rhizomania symptoms, suggesting

that resistance had been compromised. Standard soil baiting with sugar beet plants followed by ELISA tests were used to confirm the presence of BNYVV. Resistant varieties grown in regular BNYVV-infested soil remained resistant. In contrast, when grown in Imperial Valley BNYVV-infested soil all resistant varieties tested susceptible according to elevated ELISA values. Based on host reactions, eight different BNYVV isolates have been isolated from Imperial Valley soil (IV-BNYVV) by single local lesion isolation. IV-BNYVV isolates did not contain an RNA-5 as determined by RT-PCR using RNA-5 specific primers. In SSCP analyses of all the IV-BNYVV isolates, the banding patterns were identical to A-type and different from P-type. Our results indicate the resistance-breaking BNYVV isolates from Imperial Valley evolved from existing A-type (Liu, et al., 2005). In 2003 and 2004, more BNYVV-resistant breaking fields have been found in the Imperial Valley of California.

In this research, the pathogenicity of resistance-breaking BNYVV isolates in Imperial Valley, California was determined and the coat protein in RNA-2 and 25-kDa protein (encoded by BNYVV-RNA-3, involved in symptom expression) of IV-BNYVV isolates were sequenced and analyzed.

MATERIALS AND METHODS

Inoculum preparation. Each IV-BNYVV isolate was mechanically inoculated to systemic host *Beta macrocarpa,* which was planted in sterilized soil. After showing systemic infection, virus-free *Polymyxa betae* was incorporated into the soil. One month later, the infected roots and soil were used for inoculum.

Soil test. New 280 ml styrofoam cups with holes punched in the bottom for drainage were placed in sterilized plastic saucers. Cups were filled with infested soil from each isolate (one part of inoculum with nine parts of sterilized soil). A plastic divider was inserted into each pot dividing each pot into four sections. The sugar beet varieties used were rhizomania-resistant varieties: Beta 4430R (*Rz1rz1*), KWS Angelina (*Rz1rz1+Rz2rz2*) and breeding line 1927-4H5 (*Rz1rz1*+Wild beet resistance) and rhizomania-susceptible variety Beta 6600 (*rz1rz1*). Approximately 30 sugar beet seeds of each variety were layered on top of each section. Seeds within each section were covered with sand to a depth of about 1 cm. Water was added to the saucers as needed. Four replicates of each isolate were randomly placed on greenhouse benches. Each cup was about 30 cm apart to avoid contamination by splashing between cups. Greenhouses were maintained between 24-30 C. Roots from each section of these cups were harvested and tested for BNYVV by ELISA after 6 weeks post emergence of seedlings.

Root extracts preparation. Roots from each Styrofoam cup were washed free of remaining soil. Root tissue (0.2 g from each root mass) was taken from each cup and added to 2 ml of extraction buffer (0.05 M Phosphate-buffered saline, pH 7.2 with 0.5% Tween 20 and 0.4% dry milk powder). Root tissues were homogenized in sample extraction bags with a hand-held roller press (Agdia, Inc.).

Enzyme-linked immunosorbent assay (ELISA): The double antibody sandwich ELISA was used. Purified IgG made to BNYVV (1mg/ml) was used to coat microtiter plates at a 1/1000 dilution, and plates were incubated at 37 C for 1 hour. After washing 3 times with PBS-Tween (3 minutes each),

expressed sap (100 µl per well) was added to each of two wells of a microtiter plate and allowed to incubate overnight at 4C. Plates were again washed with PBS-Tween. Alkaline phosphatase-conjugated anti-BNYVV IgG was added to wells (100 µl of 1/1000 dilution). Plates were incubated for 1 hour at 37C, and then washed with PBS-Tween. Alkaline phosphatase substrate (Sigma Chemical, St. Louis, MO) were used at a ratio of 5 mg/8.3 ml of substrate buffer. Absorbance readings (A_{405nm}) were made at 1 hr after adding substrate with a Bio-Tek EL312e microplate reader (Winooski, VT). ELISA values of the test samples absorbance at A_{405nm} 3 times greater than the healthy mean was considered to be positive.

Reverse transcription-polymerase chain reaction (RT-PCR) and sequence analysis: Viral RNA extracted from purified virion preparations using the RNeasy Mini Kit (QIAGEN Inc., Valencia, CA) according to the manufacturer's instructions, was denatured by heating at 95 C for 10 min and annealed with a specific antisense oligonucleotide primer. First strand cDNA and PCR procedures were described previously (Liu, et al. 2003). The PCR products were sliced and gel purified using QIAqueck Gel Extraction Kit (QIAGEN Inc., Valencia, CA) according to the manufacturer's instructions. The eluted DNAs were sequenced by a commercial company (MCLAB, South San Francisco, CA). Sequences were analyzed by the software programs MacVector and AssemblyLIGN (Oxford Molecular Ltd., Oxford, UK).

RESULTS AND DISCUSSION

Standard soil baiting with sugar beet seedlings followed by enzyme-linked immunosorbent assay (ELISA) was conducted. Resistant varieties grown in regular BNYVV isolate-infested soil (Spence field, Salinas, California) remained resistant. In contrast, when grown in Imperial Valley BNYVV isolates-infested soil, all resistant varieties tested were susceptible according to elevated ELISA values. From the pathogenicity test, results suggested that all eight IV-BNYVV isolates could be transmited by its vector, *Polymyxa betae*, and infect all three BNYVV resistant cultivars tested (Fig. 1). Under high initial inoculum levels and optimum environmental conditions for rhizomania, disease development may appear to have breaken down partial resistant cultivars (Asher, et al., 2002). In the pathogenicity tests, we used similar amounts of inoculum and *P. betae* which will rule out the possibility that the resistance-breaking is due to the inoculum density.

The coat protein from RNA-2 and P-25 protein (encoded by RNA-3, involved in symptom expression) of IV-BNYVV isolates, were sequenced. Analyses of the deduced amino acid sequence of coat protein and P-25 protein of resistance-breaking IV-BNYVV isolates revealed the high percentage of identity with non-resistance-breaking Salinas BNYVV isolate (99.9% and >98.0% respectively). The neighbor-joining analyses (Saitou and Nei, 1987) of coat protein and P-25 protein with other known BNYVV isolates are shown in Figures. 2 and 3.

The coat protein sequence of resistance-breaking isolates and non-resistance-breaking isolates are almost identical (99.9% identity) which indicated that the resistance-breaking determinant was not on the coat protein gene.

Figure 1. ELISA Test of Sugar Beet Grown in Imperial Valley BNYVV isolates Infested Soil Compared with Salinas BNYVV Infested soil
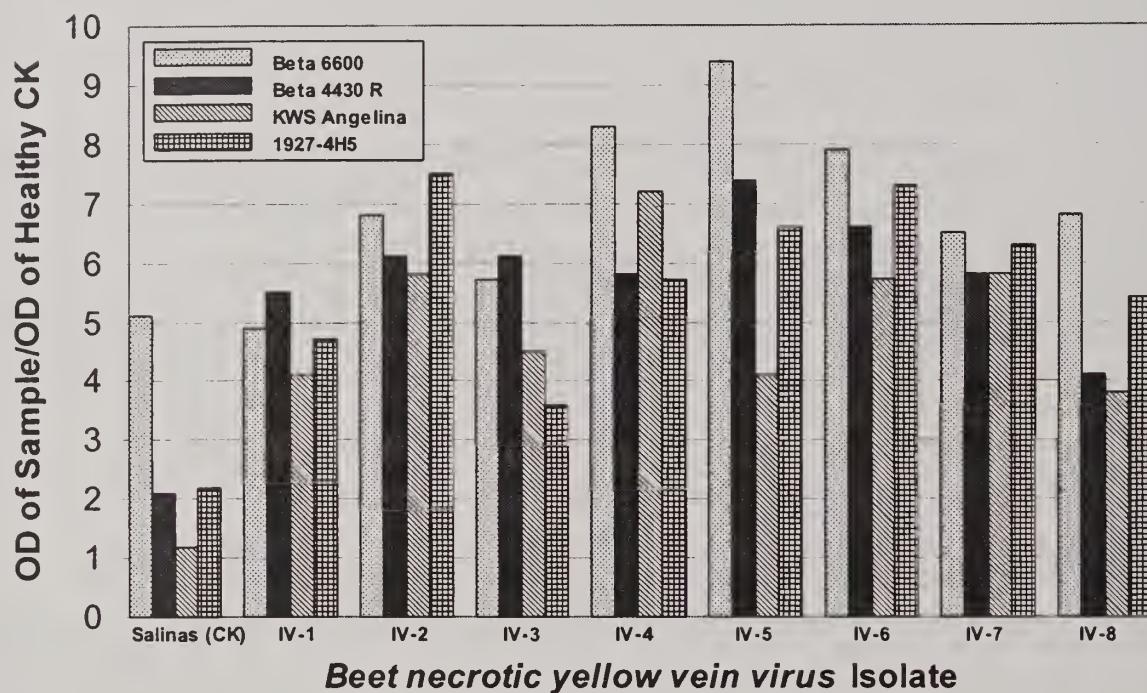
BNYVV RNA-3 facilitates the multiplication and spread of the virus in root tissue and may have a major role in the production of rhizomania symptoms. Tamada et al., 1999 reported that RNA-3 deletion mutants of BNYVV do not cause rhizomania disease in sugar beets. Single amino acid changes in the P-25 protein of BNYVV RNA-3 will determine resistance responses of *Beta vulgaris* spp. *maritima* (Chiba, et al., 2002). Nucleotide sequences for the RNA-3 encoded P-25 protein of IV-BNYVV isolates and Salinas BNYVV isolates were determined and deduced amino acid sequences were compared with each other. The P-25 proteins in all isolates consist of 219 amino acid residues and there were a maximum of 10 amino acid differences. According to the pathogenicity test, it was revealed that the difference between resistance-breaking isolates and non-resistance-breaking isolates was located at the amino acid positions 67 and 68. In order to determine whether these two amino acid changes can be resistance-breaking or not, more data is needed to draw the conclusions.

The emergence of resistance-breaking virus variants is due to genomic variation. There are four types of genomic variations in plant viruses including mutation, recombination with other RNA plant viruses, genome segment reassortment, and acquisition of a range of extra nucleic acid components (Harrison, B. D., 2002). Mutation is the most common cause of viral genomic variation and is a necessary precursor to other types of genomic variation. Among RNA viruses, the typically short replication time, high yields and high mutation rates result in virus

Fig.2. Neighbor-joining tree representing phylogenetic relationship among 27 *Beet necrotic yellow vein virus* isolates based on coat protein gene on RNA2 .

BNYVV-CP-Japan-S44
BNYVV-CP-IV-4
BNYVV-CP-Japan S42
BNYVV-CP-Iran
BNYVV-CP-Kazakhstan
BNYVV-CP-Japan-FP
BNYVV-CP-Kazakhstan-F75
BNYVV-CP-Japan-FC
BNYVV-CP-Salinas-CA-89
BNYVV-CP-IV-8
BNYVV-CP-IV-3
BNYVV-CP-IV-5
BNYVV-CP-IV-2
BNYVV-CP-IV-1
BNYVV-CP-Salinas-CA-02
BNYVV-CP-IV-7
BNYVV-CP-IV-6
BNYVV-CP-Japan-S
BNYVV-CP-Japan D104
BNYVV-CP- Japan SH1
BNYVV-CP-Iran-gp21
BNYVV-CP-France-F13
BNYVV-CP-Japan-CY3
BNYVV-CP-Japan-CH2
BNYVV-CP-China-NM
BNYVV-CP-China
BNYVV-CP-Japan-S12

87
86
78

Method: Neighbor Joining; Bootstrap (1000 reps); Tie breaking =systematic.

Distance: Poisson-correction; Gaps distributed proportionally.

cultures consisting of a complex dynamic swarm of mutants. However, most mutants are either not viable or not positively selected. The consensus sequence in such mutants will change in response to a change in environmental conditions, for example, temperature changes or continued use of resistant cultivars.

The large-scale development of resistant cultivars may impose selection pressure and lead to partial or total breakdown of resistance. Consequently, the durability of beet cultivars which are resistant to BNYVV should be assessed, not only to the original A-pathotype but also to those resistant-breaking isolates. Additional sources of resistance with different genetic determinants should also be sought to increase the stability and durability of the resistance.

Fig. 3. Neighbor-joining tree representing phylogenetic relationship among 22 *Beet necrotic yellow vein virus* isolates based on P-25 protein on RNA3.
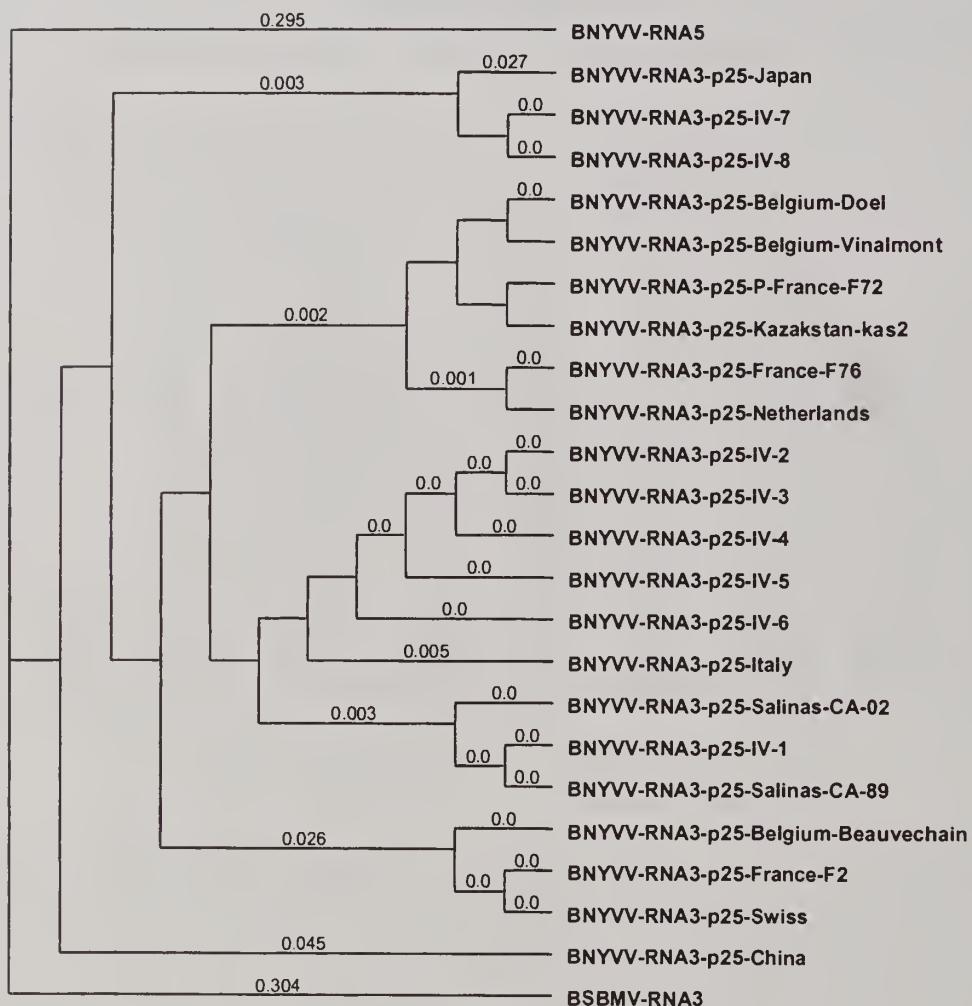
Method: Neighbor Joining; Best tree.

Distance: Uncorrected; Gaps distributed proportionally.

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Project 221

Ecology of *Beet curly top virus* (BCTV) and identification of novel sources of resistance to BCTV for bioremediation or genetic engineering

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California Beet Growers Association and California Industry Research Committee

Cooperator:
California Department of Food and Agriculture-Curly Top Virus Control Program

Introduction:

During the summer of 2001, *Beet curly top virus* (BCTV) reemerged as an important, economically damaging pathogen of sugarbeet, tomato and pepper throughout widespread areas of the western United States. These areas included California, the Snake River Valley of Idaho and the southwestern desert of west Texas and New Mexico. The disease was particularly severe in California, where extensive plant damage occurred and sugarbeet yields were reduced by several tons per acre, in large part due to losses from curly top disease (Kaffka et al., 2001). None of the sugarbeet varieties currently raised in California have any substantial resistance to curly top. Although sugarbeet varieties exist with resistance to curly top disease, these varieties are at best not capable of reaching the yields of the current varieties grown in California in the absence of curly top, and no current resistant varieties are adapted to the disease problems associated with California production. Application of systemic insecticides at planting has also been used to prevent leafhopper feeding and virus transmission, but this is only partially effective (Kaffka et al., 2002). Following the epidemic of 2001, the California sugarbeet industry began planting earlier to allow beets to reach substantial size before viruliferous beet leafhoppers moved into the fields carrying BCTV. Studies have shown that beets infected at an early age have more severe yield losses than beets that become infected later in development (Duffus and Skoyen, 1977). BCTV was also present in 2002, but beet leafhopper infestation and virus infection occurred much later and impact on the crop was minimal. Growers again planted early in 2003, but in spite of early planting, BCTV infection occurred while plants were still young. Although yields were not impacted as severely in 2003 as in 2001, infection rates were exceptionally high in 2003. This pattern will fluctuate from year to year, but the disease is constantly a production threat and will continue to impact sugarbeet production as long as there are no effective methods to significantly reduce the impact of this disease on production.

In addition to California, curly top is also a problem in Idaho and several other western states where curly top incidence and severity varies from year to year. Just as in California, curly top has been a problem in the Snake River Valley for most of the last century. In the 1920s, prior to the introduction of curly top resistant sugarbeet varieties, severe curly top epidemics resulted in 50-70 percent field abandonment in Idaho, and those fields that were harvested yielded less than 6 tons per

acre (Blickenstaff and Traveller, 1979). Even with resistant varieties, the virus is a chronic problem in Idaho and other parts of the west, including Montana, Wyoming, and occasionally Colorado, significantly affecting yields in some years.

The resurgence of curly top along with efforts to reduce and restrict pesticide usage in agriculture demonstrates a real need to develop improved control strategies for this historic and persistent pathogen of sugarbeet. Current curly top-resistant varieties, when available, do not yield as well as non-curly top resistant varieties in the absence of curly top, and this resistance is multigenic and difficult to move between varieties. Chemical control is only partially effective. With effective alternative methods for control of BCTV and curly top disease in sugarbeet, growers in all curly top affected areas could consistently plant higher yielding varieties without risk of yield loss resulting from curly top, and possibly reduce pesticide usage. Research conducted through this proposal is exploring technology-based methods to prevent infection by BCTV. Our approach is focused on development and implementation of gene silencing against BCTV using the latest technology, without genetic modification of plants. It is our intent that silencing inducers can be delivered to plants, leading to inactivation of the virus in sugarbeet plants, much as a vaccine prevents infection in humans or animals.

Objectives:

1. Develop novel genetic constructs capable of interfering with the BCTV infection process, based on current knowledge of gene silencing.

2. Transfer these constructs into a virus-based vector capable of delivering constructs to infected plants.

3. Test constructs on a model host (tobacco) and on sugarbeet to determine effectiveness of constructs in preventing virus infection.

4. Deliver constructs to sugarbeet through either genetic engineering or using a mechanical delivery system to essentially vaccinate plants against BCTV infection.

5. Complete strain identification in infected weed and crop hosts in the San Joaquin Valley.

Project Accomplishments and results from the current funding period:

Objective 1: Develop novel genetic constructs capable of interfering with the BCTV infection process, based on current knowledge of gene silencing.

Results of studies on BCTV strain identification in infected weed and crop hosts in the San Joaquin Valley demonstrated that the only two BCTV strains of significance in California are the CFH and Worland strains (see objective 5 results). Numerous variants exist that are recombinants between

these 2 strains and some variation from the original CFH and Worland isolates is present in field isolates. This variation, however, does not appear to be substantial enough to impact our proposed control method. In fact, the region of the viral genome we are targeting is highly conserved between these two viral strains. The results of our studies clearly demonstrate that CFH and Worland are the two strains that should be targeted for control of BCTV in California. Consequently, we developed contructs for virus induced gene silencing (VIGS) that should be capable of inducing silencing of both CFH and Worland strains, regardless of which would be introduced to the plant in the field.

DNA constructs were designed to target the C1 gene of BCTV. This gene is critical for virus replication and host infection, and its elimination would prevent the virus from infecting the plant. Considerable research has been conducted on both plant and animal systems that have led to an understanding of what types of genetic features trigger gene silencing. The constructs we have developed to date incorporate features shown through research to be inducers of gene silencing with both RNA and DNA viruses. Contructs were designed using the latest technology, and ordered from Invitrogen, Inc. (Carlsbad, CA), a company specializing in the manufacture of synthetic DNA for research.

Objective 2: Transfer these constructs into a virus-based vector capable of delivering constructs to infected plants.

We are using not only the virus based vector method, but also two additional approaches for delivering constructs to sugarbeet. This takes more time than focusing on a single method, but allows us to test the approach more effectively by introducing constructs to plants at different growth stages and different stages of infection. Furthermore, some delivery methods may be more effective than others, and this approach eliminates some of those variables.

We are introducing some constructs with a modified version of *Tobacco mosaic virus* (TMV), developed by a colleague and demonstrated to infect sugarbeet in our laboratory. In this approach, constructs used to initiate silencing of a gene involved in BCTV replication are incorporated individually into a specific location within the TMV genome. When TMV infects the sugarbeet plant, the target sequence is expressed as RNA and should induce a systemic signal in the plant and eliminate BCTV, preventing development of curly top disease.

In a second approach, we are introducing the constructs to plants using agroinfiltration. In this method, the same constructs are introduced individually to a piece of circular DNA in the bacterium, *Agrobacterium tumefaciens*, known as the Ti Plasmid. *A. tumefaciens* cells are grown in laboratory culture and used to inoculate plants through a process known as agro-infiltration. During this process the constructs are delivered to plant cells and expressed as RNA, which should trigger VIGS.

The final approach involves working with individual cells. We are just beginning this approach, but it has the potential to allow us to screen numerous constructs in a short period of time. This method involves the use of tobacco rather than sugarbeet cells, as a model system. In this approach, the silencing constructs, along with infectious virus DNA, are introduced directly into free floating,

tobacco cells growing in liquid culture. The cells are maintained for a period of hours, after which time they are tested for the presence of virus. If virus is present in controls (no silencing construct added) but not in test samples (both viral DNA and silencing construct added), it would suggest that the construct is effective in eliminating BCTV. We are only in the early stages of this approach, but it will allow us to screen more constructs in a shorter amount of time, with less effort than current methods. Once we identify effective constructs using this method, we can then test them in whole plant systems as described above. The problem with going directly to the whole plant system is that it requires a lot of DNA manipulation and confirmation before we can even test it in the plant, resulting in much more time and expense per construct.

Objective 3: Test constructs on a model host (tobacco) and on sugarbeet to determine effectiveness of constructs in preventing virus infection.

Initial testing of constructs began in December 2004 using the agroinfiltration method. Two approaches were used. In the first, sugarbeet plants were inoculated with BCTV-CFH strain using viruliferous leafhoppers 1 week prior to treatment with silencing constructs. The purpose of this experiment was to determine if treatment would cause the plant to bring virus under control, and would be successful if new leaves emerged on infected plants, but did not express curly top symptoms. The test has been conducted only once to date, and did not produce recovery. We acknowledge that treating plants with a construct and expecting recovery is a long shot, but if successful it would be a powerful tool for virus control, and would certainly demonstrate the effectiveness of the constructs.

The second approach is our favored method, and one for which we believe we are more likely to prevent BCTV infection. In this case, young sugarbeet and tobacco plants were treated with VIGS constructs using the agroinfiltration method, followed by inoculation with BCTV 2 to 5 days later. Initial results are promising, but much more data will be necessary to determine effectiveness of initial constructs, and additional constructs will need to be tested as well. We anticipate testing with the TMV vector constructs to begin in April. Testing with the *in vitro* (tobacco cell) system should begin approximately the same time, as we complete testing of the system itself prior to beginning construct screening.

Objective 4: Deliver constructs to sugarbeet through either genetic engineering or using a mechanical delivery system to essentially vaccinate plants against BCTV infection.

Development of field delivery systems will begin upon identification of effective constructs for control of BCTV (Objective 3).

Objective 5: Complete strain identification of infected weed and crop hosts in the San Joaquin Valley.

Sample collection. Using the extensive host range information available for BCTV, reported weed and crop hosts of the virus were collected from throughout the western and central San Joaquin

Valley using the same methods as in 2002 and 2003. Collection of weed and crop hosts of BCTV began in spring when the leafhopper vector was first becoming active and continued until October. Sample collection was conducted both by the USDA-ARS Virology Lab in Salinas, and by staff of the California Curly Top Virus Control Program (Fresno, CA). All plant samples were tested by double-antibody sandwich enzyme linked immunosorbent assay (DAS-ELISA) using polyclonal antiserum against the Logan strain of BCTV to determine which samples were infected. This antiserum effectively identifies the presence of all strains of BCTV, but cannot differentiate one strain from another.

Development of improved methods for strain identification. Last year (see 2003 report) we described the use of polymerase chain reaction (PCR)-based detection methods to identify different BCTV strains. This method involves using short strands of DNA (primers) that bind to complementary DNA sequences of a single virus strain, but not other strains. After primer binding, an enzyme extends the primers to make a copy of the original strand. This procedure is repeated several times with a set of two primers specific for a particular strain of the virus. The result can be run on an agarose gel. Samples that are closely related to the same strain as the PCR primers should have similar sequence in this region and will produce a specific band when the completed PCR reaction is run on the gel. Samples that are negative or are from a different strain should not produce this band. We have improved the efficiency of PCR-based strain differentiation, but have found that this method still does not explain the full extent of strain variation in California. Once a primer is developed and used to amplify BCTV sequence, it is necessary to periodically test the primers to be sure they amplify what we expect them to amplify (you might call this quality control). To do this we determine the DNA sequence of the PCR product and compare it to known strains and other sources of DNA available in international databases. During this process we discovered sequences that appeared to be intermediates between known strains that would amplify with primers that normally would be specific to one strain or another. As a result, we chose an alternate approach which has provided a much more complete picture of BCTV strain variation in California.

Final strain identification was based on PCR primers developed earlier in this project that amplify a 500 nucleotide section of the BCTV genome that is conserved within strains, but differs between strains. Sequence variability between strains for this section of the BCTV genome is shown in Table 1. Unlike the primers described above that amplify one strain, but not another, these primers amplify DNA from all BCTV strains ever known to be present in California. The PCR product was then run on an agarose gel as described above, along with positive and negative controls, to determine whether plants were infected with BCTV or not (for example see Fig. 1). For those crop and weed DNA samples that were positive for BCTV by PCR, we sequenced the DNA using a Li-Cor DNA analyzer recently acquired by the USDA-ARS in Salinas. Sequences from each individual plant sample were compared with known BCTV strains to determine the origin of the sequence and its nearest known relative. This method allows for identification of known strains, as well as those that differ from known strains. Results clearly demonstrated the presence of not only the expected Worland and CFH strains, but also intermediates between these strains and even a few Worland or CFH isolates with short Logan sequences interspersed (Table 2). Logan (also known as the California strain) is the strain originally associated with California. No samples from any plant were infected by the Logan strain itself. Only small segments of DNA that correspond to Logan were found interspersed between segments of CFH or Worland (identified by "L" in Table 2) and

only in a few plants. Results suggest that Logan as a strain is no longer present in California's San Joaquin Valley. The other surprising result was that a number of plants contained recombinant sequences, in which Worland and Logan sequences were both present (identified by "H" for Hybrid in Table 2). It was the discovery of such recombinants that caused us to change our strategy for strain identification. Recombinants containing small bits of Logan are by far the exception. Most recombinants contained only sequences of Worland and CFH. The majority of the strains identified were not recombinants at all, and were clearly related to one of the two predominant strains, CFH and Worland (Table 2).

Table 1. Percent identity among major curtovirus species within sequenced region.

	Worland	CFH	Logan
Logan	54%	58%	100%
CFH	64%	100%	
Worland	100%		

Figure 1. PCR of BCTV from weed samples.
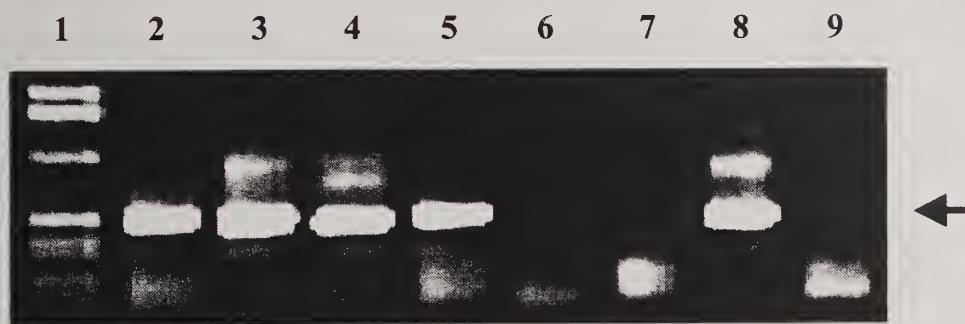
1. 1 Kb DNA Ladder
2. 03-35-12
3. 03-30-10
4. 03-37-37
5. 03-18-34
6. 03-18-4
7. 03-37-34
8. BCTV-Logan
9. Neg. Control

Most weed species were found to be infected by either individual strains, or both strains simultaneously (Table 2). Sugarbeet had by far the most diversity in strains present. Not all weed species were easily analyzed. The quality of weed material was extremely variable, and not all DNA extracted from weed species was amplifiable by PCR. PCR is a sensitive enzymatic process that can be interfered with by inhibitors that are often found in certain types of plant tissue, particularly older tissue. While not all samples collected could be analyzed for strain identity, large numbers of samples were analyzed and provided very clear results. For those species in which amplification was not possible, strain identity is listed as "ND" for Not Determined. Overall we identified slightly more CFH than Worland, but both strains are abundant in the San Joaquin Valley,

and levels appear fairly steady for each from year to year. The primary variant is the overall amount of BCTV in the valley from year to year. The years 2002 and 2004 had far less BCTV positive weed samples than 2003. This is not surprising as 2003, like 2001 was a severe curly top year.

There are a number of important conclusions from this research. First, it is clear that the Logan or California strain no longer exists in nature (at least in California). It has been replaced by the CFH and Worland strains. Previous studies by Stenger demonstrated that the origins of replication for CFH and Worland are compatible with one another, but that the Logan origin is incompatible with the other two strains. This is the section of the viral genome that we sequenced, and it is critical to the ability of the virus to reproduce itself. CFH is clearly the most aggressive of the 3 strains, particularly in crop hosts. It is possible that over time the Logan strain was unable to effectively compete with CFH (and possibly Worland) for replication factors, and as a result could not reproduce efficiently enough to be the primary virus present in a plant. Over a period of years it would have gradually diminished in prevalence to the point that it cannot be found any longer. The only remaining examples are a few pieces of sequence that have been acquired by a minor population of variants still present. Secondly, and more important for agriculture, we now know what BCTV strains are present in the field and can use this information as we develop novel approaches for control of BCTV in sugarbeet. This information is already being utilized in development of constructs discussed in the 2004 (Objectives 1-4) and 2005 proposals.

Table 2. BCTV incidence and strain identity in California weed and crop hosts: Spring 2002-Fall 2003.

Common Name	Scientific Name	+ /Total[1]	%	Strains[2]
Bassia	*Bassia sp.*	6/29	21	C, W, "L"
Bean	*Phaseolus sp.*	1/2	50	ND
Fiddleneck	*Amsinckia sp.*	0/2	0	ND
Filaree	*Erodium sp.*	1/9	11	ND
Goosefoot	*Chenopodium murale*	1/26	4	C, W, H
Hoary cress	*Cardaria draba*	0/1	0	ND
Kochia	*Kochia sp.*	0/8	0	ND
Lambsquarters	*Chenopodium album*	0/29	0	W, "L," H
London Rocket	*Sisymbrium officinale*	23/50	46	C, W
Mustard	*Brassica sp.*	32/44	73	C, W, H
Papago spinach	*Monolepis nuttalliana*	0/1	0	ND
Prostrate pigweed	*Amaranthus blitoides.*	1/21	5	C, W
Puncture vine	*Tribulus terrestris*	7/33	21	C, W
Purslane	*Portulaca oleracea*	2/16	13	W, H
Redroot pigweed	*A. retroflexus*	2/21	10	C, W
Russian thistle	*Salsola sp.*	81/141	57	C
Saltbush	*Atriplex sp.*	0/36	0	C, W
Shepherd's purse	*Capsella bursa-past.*	2/3	67	W
Sugarbeet	*Beta vulgaris*	25/32	78	C,W,"L,"H
Tomato	*Lycopersicon esc.*	26/47	55	W, C
Tree Tobacco	*Nicotiana glauca*	0/1	0	ND
Tumble pigweed	*A.graecizans*	0/9	0	ND
Wild Radish	*Raphanus sativus*	0/1	0	ND
TOTALS		**200/562**	**36**	

[1] Number positive by ELISA testing with BCTV antiserum / total number tested.
[2] C = CFH (BSCTV), W = Worland (BMCTV), "L" = Logan (BCTV)-like sequences present, H = hybrid.

Supplemental Objective: *Testing of over-wintered sugarbeet for Beet yellows virus (BYV), to determine if sugarbeet can be planted early in areas that routinely experience high levels of curly top disease.*

BYV Surveys:

As a result of the curly top epidemic that occurred in 2001 and 2003, it was in the interest of the industry to attempt early planting of sugarbeet in the spring to allow more growth and development prior to the arrival of beet leafhoppers and BCTV. It was hoped that by planting early, the industry could avoid loss of seedlings and severe stunting that can occur with early infection. On the other hand, early planting could potentially expose young beets to *Beet yellows virus* (BYV), which can also lead to significant losses for the industry. Consequently, we initiated a new testing program in

the spring of 2002 to determine the distribution of BYV in the San Joaquin Valley. By identifying areas with BYV incidence in over-wintered beet, planting could be conducted strategically. Beets could be planted early where no BYV was identified, and later in areas with BYV. This should reduce transmission of BYV from the fall crop to the spring crop in areas where BYV occurs. Areas with no BYV could be planted early to allow better growth prior to the arrival of BCTV as leafhoppers move from the foothills to the valley in late spring.

Sugarbeet leaf samples exhibiting yellowing symptoms indicative of virus yellows, a disease caused by BYV and/or two poleroviruses, *Beet western yellows virus* (BWYV) and *Beet chlorosis virus* (BChV), were collected from throughout the San Joaquin Valley by field representatives from the Mendota Factory in March 2002. These samples were brought to our laboratory, and were tested for the presence of BYV, as well as the poleroviruses, using dot blot hybridization of total nucleic acids extracted from sugarbeet leaves. Traditionally, most BYV incidence has been in the Sacramento Delta region. Interestingly, BYV was found scattered throughout the valley in the spring of 2002. Of eighteen fields tested, five were confirmed positive for BYV. Nine were positive for either BWYV or BChV. Four of the five fields with BYV incidence also had infection by either BWYV or BChV. Although the industry was more concerned about BYV than BWYV, testing for both viruses required little additional effort and provided a better picture of the types of viruses responsible for virus yellows incidence in different parts of the San Joaquin Valley. The 2004 and Spring 2005 surveys did not identify any BYV infected plants, suggesting BYV is not currently a significant threat.

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DEVELOPMENT OF SUGARBEET BREEDING LINES AND GERMPLASM

R.T. LEWELLEN

C931, C941, CR11 and CZ25/2 - C931 (PI636340), C941 (PI636341), CR11 (PI636343), and CZ25/2 (PI636342) are multigerm (MM), self-fertile (S^f), genetic-male-sterile ($A\_:aa$) facilitated, random-mated populations. These populations were developed in the population improvement program at Salinas. Very succinctly, these populations have the following relationships and attributes:

C931 = base MM, S^f, $A:aa$, $Rz1$, curly-top resistant population similar to C37/C46;
C941 = C931 x virus yellows resistant breeding lines;
CR11 = C931 x Cercospora leaf spot resistant breeding lines;
CZ25/2 = C931 x high sucrose concentration breeding lines.

C931 is a multigerm, self-fertile population. It has been under development for about 35 years. In its various developmental phases, it has been commonly used in the breeding and genetics research program at Salinas as a basic breeding population for population improvement and for selecting traits for productivity and disease resistance. C931 has the agronomic characteristics of a moderately broad open-pollinated (OP) (self-sterile) line with disease adaptation to the far western USA. However, in some breeding, selection, genetic, and germplasm improvement programs, there is a distinct advantage to be able to easily create selfed progeny families with large amounts of seed. C931 can be maintained by bulk increases or like an OP line by harvesting seed from the male-sterile segregates. The combination of self-compatibility (S^f) and genetic male-sterility allows complete flexibility in the choice of progeny and testcross families to be generated. The pedigree and development of C931 are complex and have involved both mass or mother root selection and selfed-progeny evaluation and selection. The primary source of germplasm was from C918 (PI578079) released in 1993. Thus much of the germplasm base comes from C37 (PI590715) and C46 (PI590757). Smaller portions are from C31/6 (PI590799) type sources and wild *Beta vulgaris* subsp. *maritima*. It is estimated that about 1-2% of the germplasm would have come from wild beet through C51 (PI593694).

Population C918 was the source of the *Rz1* allele that conditions resistance to rhizomania caused by *Beet necrotic yellow vein virus*. C931 is moderately resistant to *Beet curly top virus*, virus yellows caused by *Beet chlorosis virus* and *Beet yellows virus*, powdery mildew caused by *Erysiphe polygoni*, sugarbeet *Erwinia* (*E. carotovora betavasculorum*), and bolting. From C918, two cycles of S_1 progeny recurrent selection interspersed with three cycles of mass selection have been made. Germplasm from C31/6 and C51 was introduced from selected S_1 families of which C918 was a major component. One cycle of mass selection was for combined resistance to rhizomania, *Erwinia*, and slow mildewing and for sucrose concentration. The other two cycles were for resistance to rhizomania in 3-4 month old plants. For the two cycles of S_1 progeny evaluation and selection, stecklings were randomly selected and selfed. Multiple progeny tests were run at Salinas under conditions to promote moderate bolting, under nondiseased conditions, and under rhizomania

and powdery mildew conditions. Based upon nonbolting tendency, sugar concentration and yield, and resistance to rhizomania and powdery mildew, a 5-10% selection intensity was used and stecklings of the selected families were recombined through the segregating genetic male steriles. Following the final cycles of selection, two additional cycles of resynthesis were made to produce population 4931 released as C931. C931 has been tested as 8931, 9931, 0931, 1931, 2931, and 3931. C931 is an advanced sugarbeet population. It could be useful as a direct source for selecting improved parental lines. More likely, it may be most useful like it has been used in the Salinas program as an advanced population for introgressing useful traits and developing other populations and breeding lines.

C941 was developed from crosses between developmental lines of C931 and breeding lines C76-89-5 (PI593698) and C69 (PI599341). A selection and improvement program was similar to that used for C931 but greater emphasis was placed upon selecting for improved resistance to virus yellows based on virus yellows inoculated progeny tests. Because C941 is about 50% C76-89-5 and C69, its curly top resistance is less but sucrose concentration and sugar yield combining ability are slightly better. C941 has been developed and tested as 9941, 0941, 1941, 2941, and 3941. Following two cycles of resynthesis through genetic-male-sterile segregates, population 4941 was produced and is being released as C941.

CR11 was developed from crosses between developmental lines of C931 and CR09 (PI593692) and CR10 (PI593693). CR09 and CR10 have moderate resistance to Cercospora leaf spot (caused by C. beticola) derived from two Italian accessions and are 25% modern Italian germplasm. CR11 then will have about 12.5% germplasm from these Italian lines and about 87.5% germplasm similar to C931. Following one cycle of recombination, S_1 progenies were generated and evaluated for bolting tendency and resistance to rhizomania and Cercospora leaf spot. Stecklings from the selected progenies were bulked and recombined. From the recombination isolation plot, seed from individual male-sterile plants was harvested separately to create half-sib progenies. These half-sib lines were evaluated for bolting tendency, sugar concentration and yield, and resistance to rhizomania and Cercospora leaf spot. About 12% of the families were selected and stecklings from these lines were recombined to produce population CR311. Plants of CR311 were mass selected for resistance to rhizomania and resynthesized to produce population CR411 being released as CR11. CR11 has been evaluated as CR011, CR111, CR211, and CR311. CR11 should have traits similar to C931 but be substantially improved for resistance to Cercospora. In tests at Salinas, CA, Fort Collins, CO, and Shakopee, MN, its reaction to Cercospora was one grade superior to 'Monohikari' check and equal to or better than breeding line SP6822-0. Individual progeny lines expressed up to two grades better resistance than the level of the population.

CZ25/2 represents additional population improvement in population CZ25 (PI599343) released in 1997. CZ25 and CZ25/2 have about 37% of their germplasm from high sugar, $2x = 18$ lines accessed from Poland in 1988. Following one cycle of mass selection for sugar concentration and combined resistance to rhizomania and Erwinia from CZ25, individual plants were selfed to produce S_1 progenies. Progeny tests were run under bolting induction conditions and under nondiseased and rhizomania conditions. Stecklings from the selected S_1 lines were recombined with seed from each individual male-sterile plant in the pollination isolation plot harvested separately. These half-sib families were progeny tested for bolting tendency and sugar concentration and yield under both nondiseased and rhizomania conditions. Based upon nonbolting

tendency and sugar concentration and yield, stecklings from approximately 12% of these progeny lines were resynthesized to produce population Z325. Z325 was reselected for resistance to rhizomania and again resynthesized to produce population Z425 being released as CZ25/2. CZ25/2 has been evaluated as populations Z125, Z225, and Z325. CZ25/2 has improved sugar concentration compared to C931 and based upon extracted progeny lines, selection for high sucrose in combination with fair to moderate resistance to curly top and virus yellows is possible.

Populations C931, C941, CR11, and CZ25/2 should be useful as sources of combined disease resistance with good potential for productivity. Direct extraction of potential parental lines may be possible.

Seed of C931, C941, CR11, and CZ25/2 will be maintained at the USDA, ARS, U.S. Agricultural Research Station, Salinas, California, and will be provided upon written request to sugarbeet breeders in sufficient quantities for reproduction. Genetic material of these releases has been deposited in the National Plant Germplasm System where it will be available for research purposes, including development and commercialization of new parental lines and cultivars. It is requested that appropriate recognition be made if this germplasm contributes to the development of a new breeding line or cultivar. The National Germplasm System and additional information on prior releases and PI numbers can be found at: www.ars-grin.gov.

CN12 and CN72 - CN12 (PI636338) and CN72 (PI636339) are multigerm (*MM*), self-fertile (S^f), genetic-male-sterile (*A_:aa*) facilitated, random mated populations that segregate for resistance to sugarbeet cyst nematode (SBCN) (*Heterodera schachtii*). Based upon greenhouse tests of individual plants, both populations have about 40% of their plants that are moderately to highly resistant to SBCN. The resistance factors were derived from different wild beet (*Beta vulgaris* subsp. *maritima*) accessions but these accessions may have initially come from the same or similar source. Inheritance of resistance has yet to be determined, but empirical results and performance of selections, progeny lines, and hybrids suggest that resistance is highly heritable, dominant, and due to one or a few genetic factors.

CN12 is a multigerm, self-fertile population that segregates for genetic male sterility. CN12 segregates for resistance to sugarbeet cyst nematode, powdery mildew (*Erysiphe polygoni*) conditioned by *Pm*, and rhizomania (*Beet necrotic yellow vein virus*) conditioned by *Rz1*. Most, if not all, of the annualism (*B*) of the wild beet ancestry has been eliminated and CN12 has moderate nonbolting tendency. It has moderate resistance to *Beet curly top virus*, virus yellows (*Beet chlorosis virus* and *Beet yellows virus*), and sugarbeet *Erwinia* (*E. carotovora betavasculorum*). Its developmental lines have shown good yield performance particularly under natural infection with SBCN and rhizomania. The precise frequency of resistance to SBCN has not been determined nor has the efficacy of this resistance been fully characterized in field and greenhouse tests.

Theoretically, CN12 is 12.5% wild beet with about equal proportions of wild beet-97 (PI546394) and wild beet-242 (PI546413). WB97 and WB242 were each crossed to breeding line C54 (PI590802). In 1991, these F_1's were combined and crossed to genetic-male-sterile plants from population 0747 (PI590762). Population 0747 is similar to C37 (PI590715) and one of the progenitors of population C931 (PI636340). Plants within the F_1BC_1 generation were selected

under field conditions for resistance to powdery mildew and increased in bulk to produce line P202. Line P202 was grown in the field in 1993 under natural powdery mildew and SBCN conditions. When individual plants were examined and selected, it was observed that in addition to segregating for reaction to powdery mildew (Pm-:$pmpm$), some roots were heavily infested with SBCN cysts and a few were completely free of visible cysts. The selected plants were divided into two groups, one that was free of cysts to become P402NR and one that had high resistance only to powdery mildew to become P402. P402 and P402NR were crossed to population C931.

Individual F_1BC_2 families were grown under naturally infected rhizomania and powdery mildew conditions at Salinas in 1997. Plants dually resistant to rhizomania and powdery mildew from all families were combined and increased in bulk to produce P812. A second cycle of mass selection for resistance to powdery mildew, rhizomania, freedom from bolting, and agronomic type was done to produce P912. Up to line P912, selected plants had been increased in bulk in isolation and could have produced various combinations of sib matings and selfs to produce mixtures of S_0, S_1, and S_2 plants. P912 would have likely segregated for self-fertility, genetic male sterility, powdery mildew, rhizomania, hypocotyl color (R:rr), etc. Population P912 was selected by mass selection two additional times in the field at Salinas under natural rhizomania, SBCN, and powdery mildew conditions to produce lines called N112 and then N312 in 2003. In addition to resistance to diseases, roots were selected on the basis of agronomic type, size, and sugar concentration. Selection for resistance was done visually at harvest with only plants resistant to all three diseases selected, but it is likely that escapes, particularly for SBCN, occurred under these field conditions.

In addition to mother root selection and bulk increase, individual plants from population P912 were selfed to produce N112-# progenies. These selfed progeny families were evaluated for performance at Salinas and Brawley under both diseased and nondiseased conditions. At Brawley, the progeny test was under severe rhizomania and SBCN conditions. Based upon line performance in these tests, individual stecklings from within the selected progenies were bulked and increased by selfing to produce a second cycle of selfed progeny families called N212-#. The second cycle progenies were also evaluated at Brawley and Salinas in a series of tests that evaluated performance, bolting tendency, and disease resistance. From 48 second cycle families, stecklings from 14 families were selected and bulked. This bulk was combined with stecklings from N312 in approximately equal proportions and recombined through the segregating genetic male steriles to produce population N412 released as CN12.

CN72 is a multigerm, self-fertile population that segregates for genetic male sterility. It has approximately 25% wild beet germplasm. CN72 segregates for resistance to SBCN and rhizomania conditioned by $Rz1$. Most of the annualism of the wild beet ancestry has been eliminated. The precise frequency of resistance to SBCN has not been determined. It is not known if the resistance in CN72 is identical to that in CN12. The wild beet source of resistance to SBCN was a Salinas accession from Europe that had been reported to be tolerant/resistant to SBCN. The increase of this accession at Salinas in 1994 was called N499 (PI599349). In 1997, plants from N499 were crossed to genetic male sterile plants from population C931. F_1 plants selected for resistance to rhizomania were backcrossed to C931 to produce line N972. N972 was developed in parallel with P912 (see above). The same criteria of selection were used except N972 does not segregate for Pm that conditions high powdery mildew resistance. Identical kinds of lines and selfed progenies were produced. After two cycles of mass or bulk selection and increases, lines N172 and then N372 were

produced. Similarly, after two cycles of selfed family selection from N972 progeny lines N272-#s were produced. Because N972 would have segregated for genetic male sterility and self-fertility and pollination was not controlled within the isolation chambers, N372 could have been composed of S_0, S_1, and/or S_2 plants. Stecklings of N372 and from 10 selected families out of 24 of N272-# S2's were combined in approximately equal numbers and recombined through the genetic male steriles to produce N472, released as CN72.

CN12 and CN72 are being released as possible sources of resistance to SBCN in enhanced backgrounds. Selections from CN12 may lead to potential parental lines but CN72 retains too many wild beet influences for this purpose. Both lines will likely need to be further backcrossed to advanced sugarbeet parental lines to increase components of sugar yield and eliminate sprangling. These lines may be useful for biological and agronomic tests to evaluate the efficacy of these sources of SBCN resistance and to establish progenies to search for molecular markers.

Seed of CN12 and CN72 will be maintained at the USDA, ARS, U.S. Agricultural Research Station, Salinas, California, and will be provided upon written request to sugarbeet breeders in sufficient quantities for reproduction. Genetic material of these releases has been deposited in the National Plant Germplasm System where it will be available for research purposes, including development and commercialization of new parental lines and cultivars. It is requested that appropriate recognition be made if this germplasm contributes to the development of a new breeding line or cultivar. The National Germplasm System and additional information on prior releases and PI numbers can be found at: www.ars-grin.gov.

EVALUATION OF CULTIVARS OF SUGARBEET WITH CYST NEMATODE RESISTANCE IN CALIFORNIA

- Sugarbeet cyst nematode (SBCN) (*Heterodera schachtii*) can be a very damaging pest of sugarbeet (*Beta vulgaris*). At different times and locations, it has been considered problem number one. Much research has been expended to study the biology of SBCN, to determine control practices, and to find host-plant resistance. SBCN has necessitated the use of crop rotations, chemical protectants, and soil fumigants and has been responsible for the dislocation of production areas, usually away from factory sites. Worldwide breeding for resistance to SBCN in sugarbeet has been an objective for about 90 years. At Salinas, since about 1950, breeding for resistance to SBCN has been a primary objective. Scientists who have worked on the biology and control of SBCN and resistance in sugarbeet at Salinas have included Mr. C. Price and A.E. Steele and Drs. A.M. Golden, H. Savitsky, D.L. Doney, E.D. Whitney, J.S. McFarlane, M.H. Yu, and more recently, R.T. Lewellen. Extensive research has also been done by the University of California at Riverside and Davis. In the 2003 California Sugar Beet Annual Report, California Beet Growers Association, Dr. M.H. Yu reviewed the history of the worldwide breeding efforts to produce sugarbeet cultivars that are highly resistant to SBCN.

In summary, within cultivated beet, *B.vulgaris*, high resistance has not been found. In the progenitor of all cultivated beets, *B.vulgaris* subsp. *maritima*, tolerance and partial resistance has been reported. The highest level of resistance was identified in the *Patellares* section of *Beta* in the hard seeded species *B.procumbens*, *B.webbiana*, and *B.patellaris*. Because of the near-immunity of resistance in these hard seeded species, this resistance has received the most attention of genetic and breeding efforts. Recently in Europe, hybrid cultivars with resistance to SBCN from *B. procumbens* conditioned by the gene Hs^{pro-1} have been marketed on a limited scale. In 2004, experimental

hybrids from Betaseed and Syngenta (Hilleshog) and from the USDA program at Salinas with resistance from *B.procumbens* were evaluated in tests at Brawley and Salinas (Table 1).

As mentioned, partial resistance has been known in one or a very few accessions of *B. vulgaris* subsp. *maritima*. For example, H. Reitberg in the Netherlands identified from the Loire Valley Estuary in France specific collections that had partial resistance. The best known of these is now known as Wild Beet 242 (WB242) and is stored in the national seed bank as PI546413. There now also may be other sources of resistance identified and being used from within the *B.vulgaris* – *B.vulgaris* subsp. *maritima* continuum. These *maritima* and/or other sources of resistance have received renewed interest and breeding activity in USDA-ARS research programs at Fort Collins, CO and Salinas, CA and by commercial seed companies. Breeding lines CN12, CN72, and C927-4 have recently been released from the program at Salinas. Based upon field and greenhouse tests, these lines segregate for high levels of resistance to SBCN. The resistance in CN12 is believed to be from WB242. It is less clear where the resistance in CN72 and C927-4 was derived and whether it is different from the resistance in CN12. In tests at Salinas and Brawley in 2004, these breeding lines and two experimental hybrids from Betaseed with non-*B.procumbens* resistance to SBCN were evaluated (Tables 1, 2, & 3).

The tests in 2004 at Brawley and Salinas of the nematode resistant varieties and lines were grown under both SBCN infested and noninfested conditions. Where there was SBCN infestation, the soil was infested also with rhizomania (*Beet necrotic yellow vein virus*). In the SBCN tests, there were 24 varieties in 2-row plots with eight replications. The results of only some of these are shown (Tables 1, 2, & 3). Soil cores from eight varieties were taken for SBCN counts. Cores were taken 12 inches deep, 3-4 inches from the plants on the inside shoulder of the beds. Eight random cores were taken per plot and combined into one sample per plot. Nematode counts were made by L.Pakish at Salinas. For the Brawley test B504, soil was sampled 03/04/04 and at harvest 05/23/04 (Table 2). Samples for initial SBCN population counts were not made. At Salinas, the trial 3504 was sampled three times: early in the season on 06/01/04; midseason on 09/03/04; and at harvest at 11/05/04. The trials were harvested and root yield and sugar content determined. At Brawley, the sugars were determined by Spreckels Sugar. The test B504 at Brawley was under high SBCN pressure and mild rhizomania. An adjacent test B304 under similar cultural conditions was nearly disease free and was used to calculate relative sugar yield between diseased and nondiseased conditions (Table 2). At Salinas, in test 3504, rhizomania was considered to be more severe than the SBCN infestation. Sugar yield comparisons are given in Table 3 between the same varieties in tests 3504 and nondiseased test 704. At harvest on 11/23/04 for test 3504, the roots were lifted, scored for rhizomania on a scale of 0 to 9 where 9 = very severe or dead, bagged, washed, weighed, and run through the sugar lab. For rhizomania, roots scored 0-5 were considered resistant (Table 3). Based upon the 2004 test results at Brawley and Salinas, combined resistance to SBCN and rhizomania is happening. Under high nematode populations at Brawley, it appears that both resistance from *B.procumbens* and the other sources condition resistance. The interesting and highly encouraging finding is that the resistance from the other sources appears to be as efficacious at protecting potential sugar yield and reducing nematode buildup as the *B.procumbens* source. Under lower populations at Salinas, the results are less dramatic, but the final SBCN counts were reduced by both kinds of resistance (Table 3).

The lines evaluated from the Salinas breeding program appeared to be intermediate to the results from the commercial hybrids. Tests in the greenhouse on individual plants showed that CN12, CN72, and C927-4 segregate for reaction to SBCN. Within each source, highly resistant plants occur. Evaluation, screening, and selection within these lines and others should produce homogeneous resistant lines. It is believed that when these reselected lines and their hybrids are tested, that their nematode resistance will be highly effective.

These test results based upon only one year's data need to be considered preliminary. The tests are being repeated in 2005 at both Brawley and Salinas. In addition to the Syngenta and Betaseed hybrids, nematode resistant hybrids from Holly Hybrids also will be evaluated. The Betaseed hybrid tested as 2AP0852 has been provisionally accepted for limited marketing by the Imperial Valley Seed Committee as 'Beta 8520N.' After so many decades of work, it is exciting to think that resistance to SBCN is now approaching commercial reality.

Table 1. Hybrids and breeding lines evaluated for resistance to sugarbeet cyst nematode (SBCN) at Brawley and Salinas, CA in 2004.

Variety	Resistance			Description
	Rz	NR1	NR2	
Beta4430R	√			Betaseed commercial hybrid
Phoenix	√			Holly Hybrids commercial hybrid
US H11				Susceptible check
Hil-1	√	√		Syngenta hybrid (Hilleshog)
Hil-2	√	√		Syngenta hybrid (Hilleshog)
Hil-3		√		Syngenta hybrid (Hilleshog)
2VK0305	√	√		Betaseed hybrid
0VK6280		√		Betaseed hybrid
2AP0852	√		√	Betaseed hybrid 'Beta 8520N'
2EN5066			√	Betaseed hybrid
CN12	√		√	USDA-ARS breeding line
CN72	√		√	USDA-ARS breeding line
C927-4H50	√		√	USDA-ARS experimental hybrid

Rz = rzm resistance due to $Rz1$ or other sources of resistance.
NR1 = *Beta procumbens*, Hs^{pro-1} nematode resistance.
NR2 = *Beta vulgaris* subsp *maritima* or other sources of resist.

Table 2. Performance at Brawley of hybrids and breeding lines with resistance to SBCN under high SBCN and mild rhizomania conditions versus low SBCN populations in 2004.

Variety	Test B504 with SBCN			Test B304 w/o SBCN	
	Sugar Yield lbs/a	Eggs & Larvae (no./100 g soil)		Sugar Yield lbs/a	Relative %
		3/4/04	5/23/04		
Beta 4430R	4900	10590	12420	13500	36
Phoenix	4500	12720	16220	12200	37
US H11	2300			8900	26
Hil-1	8000			9700	83
Hil-2	8800	1410	2350		
Hil-3	7700				
2VK0305	10400	1870	3220	11400	91
0VK6280	6700				
2AP0852	10800	1710	2651	12200	89
2EN5066	9000				
CN12	6200	5540	6630	9900	62
CN72	5800	6490	5510	8200	70
C927-4H50	6900			12600	55
LSD (.05)	770	2650	3740	1450	

Test B504 planted 9/15/03 and harvested 5/25/04.
Test B304 planted 9/15/03 and harvested 5/21/04.

Table 3. Performance at Salinas of hybrids and breeding lines with resistance to SBCN under moderate SBCN and severe rhizomania (RZM) versus Nondiseased conditions in 2004.

Variety	Test 3504 with SBCN					w/o SBCN
	Sugar Yield lbs/a	RZM %R	Eggs & Larvae (no./100 g soil)			Sugar Yield lbs/a
			6/1/04	9/3/04	11/5/04	
Beta4430R	14100	95	105	314	715	17800
Phoenix	13400	84	208	544	387	16000
Hil-1	11400	89				14700
Hil-2	11900	82	269	79	73	14100
Hil-3	7200	50				15000
2VK0305	14000	85	216	147	49	16300
0VK6280	7600	58				15200
2AP0852	14600	90	148	130	63	15400
2EN5066	10600	78				14600
CN12	11700	78	294	139	193	15100
CN72	11700	62	288	106	209	14200
C927-4H50	13100	85	290	170	491	17000
LSD (.05)	970	8	NS	171	244	1390

Test 3504 planted 4/26/04 and harvested 11/23/04.
Test 704 planted 3/19/04 and harvested 9/29/04.

EVALUATION OF GERMPLASM TO IV-BNYVV STRAINS AND BREEDING FOR RESISTANCE

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(Note: This is a portion of a feature article for publication in
Plant Disease in collaboration with C.M. Rush, et al.)

Sources of resistance and their inheritance –
After rhizomania was originally found in the USA (Duffus et al., 1984), the USDA-ARS's breeding program at Salinas, CA did an extensive search to identify genetic variability for reaction to BNYVV, to find sources of resistance, and to develop resistant and enhanced germplasm and breeding lines (Biancardi et al., 2002). The so-called "Holly" resistance or "Holly" gene was found in 1983 by A.W. Erichsen at Tracy, CA in the Holly Sugar Company breeding program and was shown to be inherited as a single dominant major gene (Lewellen, Skoyen, Erichsen, 1987) and named *Rz1* (Lewellen, 1988; Scholten et al. 1999). Other than *Rz1* and the resistance in the cultivar Rizor developed by SES in Italy (Buttner et al. 2004; DeBiaggi, 1987; Biancardi et al., 2002) no other major gene resistance was identified within sugar beet (Biancardi et al, 2002; Scholten et al., 2000).

However, it was apparent in field trials that different degrees of susceptibility or tolerance (resistance) occurred within sugar beet breeding lines. Lines C39 and C47 were more tolerant than most other breeding material as measured by symptoms and sugar yield (Lewellen, 1995b; Lewellen et al., 1987; Lewellen and Biancardi, 1990). Recurrent phenotypic selection based on the visual reaction and yield of individual plants under severe disease conditions in the field were used to make improvements. After five cycles of selection, breeding lines C39R and C47R were developed (Lewellen, 1995b; Lewellen & Biancardi, 1990). Based upon disease symptoms and yield selection under severe rhizomania conditions, the level of resistance in C39R and C47R was equal to that conditioned by *Rz1* (Lewellen and Biancardi, 1990). This resistance was quantitative or additive and was transmitted to experimental hybrids to an intermediate (mid-parent) level compared to the dominant *Rz1* allele. This C39R type resistance reduced symptom expression (Table 1), but did not reduce BNYVV titer in infected roots (Lewellen & Biancardi, 1990) and has been little used. The *Rz1* allele was much easier to control and follow in breeding programs and lent itself to rapid deployment in backcrosses and population improvement programs using either visual symptom expression, ELISA values for BNYVV, or marker assisted selection (Scholten et al., 1997; Francis et al., 1998; Pelsy and Merdinoglu, 1996). Quickly, *Rz1* became the favored resistance against rhizomania worldwide. Among breeders though, it was observed that the high, but partial resistance of *Rz1*, did not perform equally in all backgrounds or hybrid cultivars. It is thought that the *Rz1* cultivars with the best expression of resistance may have resistance supported from quantitative or minor gene additive resistance as found in C39R and C47R. It is known also that *Rz1* acts in a partially dominant manner and that *Rz1* allelic dosage in diploid versus triploid hybrid cultivars affects disease expression (Wisler et al., 1999) and that other soil borne viruses interact in a competitive or synergistic manner in sugar beet (Wisler et al., 2003).

With the knowledge that single dominant resistance genes are frequently vulnerable to loss due to changes or selection pressure to virulence within the pathogen, the search for additional resistance

genes was not abandoned. With the lack of further success within sugar beet itself, the search for resistance was quickly expanded to the larger *B.vulgaris* germplasm resources and particularly to *B.vulgaris* subsp. *maritima*, the ancestral form of all cultivated beet. Two general breeding approaches were used. One was to target specific individual accessions or plant introductions (PI's) and the other was to search within pooled or composited collections. In the first, when resistance was found within a specific accession, that resistance was backcrossed into sugar beet breeding lines. Resistance was identified in a number of accessions in field (Lewellen, 1995a; 1997) and greenhouses tests with ELISA (Whitney, 1989). For example, the resistance found in *B.vulgaris* subsp. *maritima* accession WB42 was crossed into sugar beet breeding line C37 (Lewellen & Whitney, 1985) and was released as C48 (Lewellen & .Whitney, 1993) and C79-3 (Lewellen, 1997) (Table 1). Subsequently, the resistance from WB42 was shown to be at a different locus than *Rz1*, to condition a higher level of resistance in growth chamber tests, and was named *Rz2* (Scholten et al., 1996; 1999; Buttner, 2004). Continuing research has shown that most of the identified sources of resistance are *Rz1* or *Rz2*, but every case has not been determined (Biancardi et al., 2002).

In the second approach, collections of *B.vulgaris* subsp. *maritima*, for example, the collections of Doney et al. (1990), were individually screened for resistance to rhizomania, but selected plants were pooled and increased in mass. For example, populations that lead to C26, C27, and C51 (Lewellen, 2000); R21; C67/2 (Lewellen, 2004); and R23, R23B, and R20 (Lewellen, 2000) were produced (Table 1). These very broadly based germplasm populations that had been improved for resistance to rhizomania and sugar beet traits were among the lines evaluated in 2004 for reaction to the IV-BNYVV strain of rhizomania (Table 1). For these populations, no attempt at Salinas had been made to determine if resistance was due to *Rz1*, *Rz2* or other factors. So by 2002, when the IV-BNYVV strains appeared, an extensive germplasm base with resistance to rhizomania had been developed.

Breeding activities - In 2002 when it appeared that a new strain or race of BNYVV had developed or had been selected out of the genetic diversity of BNYVV types (Liu et al., 2005), this inventory of previously developed germplasm with resistance to rhizomania was one of the most logical places to search for additional resistance factors. Tests were run in the greenhouse and field at Salinas and Brawley, CA. In those tests, the hybrid cultivars Beta4430R and Angelina were used as checks (Table 1).

The aggressive P-strain in France was known to cause significant damage against cultivars with only *Rz1* resistance (Koenig and Lennefors, 2000; Büttner et al., 2004; Richard-Molard, 2002; Harju and Richard-Molard, 2002). The hybrid cultivar Angelina developed by KWS SAAT AG in Germany was shown to have better resistance against P-strain (Harju and Richard-Molard, 2002; Büttner et al., 2004). Angelina possesses both *Rz1* and *Rz2* for resistance to BNYVV (Harju and Richard-Molard, 2002). The cultivar Beta4430R has the genotype *Rz1rz1*, *rz2rz2*. Fully susceptible cultivars such as USH11, Beta6600, and Roberta are *rz1rz1*, *rz2rz2*. Combinations of these cultivars have been used at Salinas as checks to measure ELISA values of BNYVV baited plants from greenhouse soil tests and for yield and disease scores in infested field tests (Table 1). Based upon ELISA values, *Rz1* protected against normal A-strain as represented by soil from the Salinas trial fields but not against IV-BNYVV from Imperial Valley fields Rockwood 158 and 156 where the first case of defeated resistance had been observed (Liu et al, 2005). Angelina under

Rockwood soils in greenhouse tests tested positive for BNYVV, but the ELISA value was usually intermediate, suggesting a partial or moderate resistance to IV-BNYVV.

In 2003-2004, a modified extension of the greenhouse baiting tests were run to evaluate sugar beet lines for reaction to BNYVV. Either sterilized Salinas soil, BNYVV infested Salinas soil, or Rockwood 158 (IV-BNYVV) soil was mixed with sterilized river sand in a 1:9 ratio and placed into 30x60 cm seedling flats. Ten rows of seed were sown into each flat in random order and each set of line x soil treatments was replicated three times. At 6 weeks post-emergence, roots from all seedlings within each treatment were combined and tested by ELISA for BNYVV. Some of these data are summarized in Table 1. Briefly, under sterilized soil conditions, all entries were negative for BNYVV. Under Salinas BNYVV conditions, only the susceptible checks scored positive (3x higher than healthy check). Under IV-BNYVV soil (Rockwood 158), no variety tested resistant (<3x the ELISA value of the healthy check) but, Angelina and breeding lines with *Rz2* and/or *Rz1* & *Rz2* were intermediate in ELISA values as compared to entries with only *Rz1* or no resistance allele. In general, these results matched the results from the tests run by Liu et al. (2005) and the subsequent field tests at Hartnell. Test results were confounded, however, by most of the entries not having known allelic frequencies. In the case of the *Rz2* backcross derived lines, e.g., C79-3, the resistance allele probably occurred at less than 50%. Any previously unknown resistance alleles could be at even lower frequencies and masked by the preponderance of susceptible plants.

No conclusion could be made from the greenhouse seedling tests whether there had been new resistance genes against IV-BNYVV. Even if a low frequency of highly resistant plants occurred, they could not be detected and selected by this method. On the other hand, field tests under IV-BNYVV conditions would allow large numbers of plants from many divergent germplasm sources to be screened and individually selected for resistance. As needed, follow-up progeny tests and re-evaluations with ELISA could be run to determine if selected individuals were truly highly resistant or escapes. This procedure was known to work efficiently for the original screening for resistance to rhizomania (Biancardi et al., 2002). Rare resistant individuals could be found among thousands of screened plants. This procedure requires evaluation in field plots in uniform disease nurseries. In 2003, a disease nursery was established at Salinas in an isolated location (Hartnell field). In Imperial Valley, Rockwood 156, which showed disease in 2002-2003, was chosen as a location for field tests in 2003-2004. For future trials in the Imperial Valley, an infested trial field is being developed at the Imperial Valley Research Center, Brawley, CA.

In September 2003, a coded variety trial was sown in the Rockwood 156 field that had shown rhizomania in 2002-2003. This trial was grown under near commercial conditions. The purpose of the trial was to allow advanced experimental hybrids to be evaluated under IV-BNYVV conditions. Part of the results of this coded variety trial is summarized in Table 2. Although the entry pedigrees and resistance factors were maintained as proprietary, it was known that these hybrids represented various combinations of resistance factors. There was a wide dispersion of means for sugar yield and root rot due to rhizomania. Compared to two widely grown *Rz1* commercial hybrids Beta4430R and Beta4776R, there were moderately resistant entries. However, compared to ongoing commercial harvest at this time under non-IV-BNYVV conditions, it was still obvious that there is a long way to go to prevent yield loss and root rot against IV-BNYVV. The expressed consensus of the breeders was that whether with *Rz1* or *Rz2* resistance, minor genes were very important in the field performance of these hybrids.

For the Hartnell field, as far as could be determined, sugar beet had never been grown and the field was isolated by more than 100km from the nearest commercial sugar beet production. In June 2003, IV-BNYVV inoculum (viruliferous *P.betae*) that originated from Rockwood 158 was incorporated into the soil and an *Rz1* cultivar grown to encourage the increase of IV-BNYVV strains. In this inoculation crop, root symptoms were very mild, but plants tested positive for BNYVV by ELISA and a very low frequency of systemic infection was observed. Liu (unpublished) showed that BNYVV from the Hartnell field fit the molecular profile of IV-BNYVV strains. Check varieties Z210H50 (susceptible), Beta4776R (*Rz1*), Beta4430R (*Rz1*) and Angelina (*Rz1,Rz2*) tested as root composites in September 2003, had ELISA values of 8.2, 9.4, 10.4, and 2.9 compared to 1.0 for healthy check. In October, this inoculum crop was incorporated into the soil.

In May 2004, after the soil was warmer than 16°C, sugar beet trials were established. Three tests or types of material were grown. Entries in one test were breeding lines and populations that represented a wide sugar beet germplasm base. In a second set of materials, entries were populations and lines that had a recent history of being partially derived from *B.vulgaris* subsp. *maritima* or were wild beet populations. In November 2004 from these two sets of materials, the plants were lifted and based upon visual root symptoms, size, shape, and sucrose concentration, purportedly resistant plants were selected and placed in cold rooms for vernalization for seed production in 2005. ELISA tests were not used to make these selections because it is known that from late fall harvested roots from the field, ELISA tests for BNYVV are highly unreliable. The results of these selections will not be known until their progeny are evaluated starting in 2005-2006.

The third test in 2004 in the Hartnell field was designed to screen a wide array of lines and germplasm for performance under IV-BNYVV conditions. The purpose was to try to identify among and within breeding lines and germplasm sources, potential sources of resistance and the efficacy of this resistance. At harvest in November 2004, the plots were lifted and each beet scored for rhizomania. The roots of each plot were then topped, placed into bags, washed, weighed, and run through the sugar analysis laboratory. After lifting, the roots were scored visually for rhizomania on a scale of 0 to 9 (Wisler et al., 1999), where 0 and 1 were considered highly resistant. A disease index (DI) was calculated for each plot and percent resistant (%R) roots calculated where only 0 and 1 were considered resistant. Results for some of the selected entries are summarized in Table 1.

Based upon the preliminary field trials, definitive conclusions can not be drawn. The field results did largely support the greenhouse results using ELISA. The *Rz1* resistance was defeated but under the conditions of these tests appeared to continue to provide some protection. The *Rz2* resistance either with or without *Rz1* appeared to condition moderate resistance. Neither of these factors appeared to be sufficient to give high levels of resistance and prevent damage and yield loss in the field. Individual plants in some breeding lines tested as highly resistant and the frequency of these differed among entries. Until follow-up tests are run on the apparently highly resistant individuals selected, it will not be known if they represent disease escapes, various combinations of *Rz1* and/or *Rz2* with modifiers, or if new resistance genes or alleles have been identified. One thing is obvious, with the variability in BNYVV and for the fairly rapid emergence of resistance breaking strains, sugar beet breeders will be challenged to stay ahead of the disease.

Table 1. Screening for sources of resistance to IV-BNYVV in greenhouse and field tests, Salinas, 2004

Cultivar or Line	Description (Reference)	Genes[a]	Greenhouse ELISA value[b]			Hartnell field[c]		
			H	BNYVV	IV-BNYVV	DI	%R	SugarYld kgha[-1]
Hybrids								
US H11	susc. check	---	1.0	4.6	12.4	4.7	10	4700
Beta6600	susc. check	---	1.0	6.5	13.8			
Roberta	susc. check	---				5.0	4	6500
Phoenix	commercial	*Rz1*				3.7	25	7700
Rizor	commercial	?				4.3	11	7300
Beta4430R	commercial	*Rz1*	1.0	1.1	12.8	2.6	48	9600
Angelina	commercial	*Rz1,Rz2*	1.0	1.3	4.6	2.3	49	11400
Breeding lines								
03-SP7622-0	susc. check	---				5.1	3	3300
01-EL0204	( )	*Rz1*				2.5	42	7900
C69/2	( )	*Rz1*				3.4	20	9600
C39R	( )	quant.				2.6	33	11200
C47R	( )	quant.				2.9	34	10700
02-US22/3	susc. check	---				4.8	3	5100
C37	( ),susc. check	---				3.8	9	5400
C79-2	( ),C37xWB41	*Rz2*				3.4	18	6300
C79-3	( ),C37xWB42	*Rz2*	1.0	0.9	8.3	3.4	13	6500
C79-8	( ),C37x*Bvm*	*Bvm*				3.5	21	6200
C79-C	C37 x *Bvm*	*Rz2,Bvm*				2.9	33	8400
Sugar beet x *B.vulgaris* subsp. *maritima*								
C26	( ),C37x*Bvm*	*Bvm*				3.2	32	7300
C27	( ),C69/2x*Bvm*	*Rz1,Bvm*				2.9	37	8000
R21	C26xC27	*Rz1,Bvm*	1.0	1.0	11.9	3.3	23	7600
C67/2	( ),C78xC51	*Rz1,Bvm*	1.0	1.0	9.0	3.0	31	9000
B.vulgaris* subsp. *maritima								
R423	( ),*Bvm* comp	*Bvm*				3.2	30	------
R423B	( ),*Bvm* comp	*Bvm*				2.8	36	------
R720	( ),*Bvm* comp	*Bvm*				3.3	32	------
LSD (.05)			2.8	2.8	2.8	1.1	20	1900

[a]Quant = quantitative resistance without known major gene resistance. *Bvm* = resistance from *B. vulgaris* subsp. *maritima* but nature of resistance factors is unknown.

[b]Roots from baited seedling plants grown in flats where H = healthy soil (sterilized), BNYVV = soil from Salinas rhizomania trial field, and IV-BNYVV = soil from Rockwood 158, Imperial Valley.

ELISA values = ratio of the absorbance at 405nm reading for sample over corresponding healthy absorbance value. Ratio of ≥3 x the healthy mean are considered infected.

[c]Sown as 96 entry x 4 reps, RCB on May 5, 2004; harvested November 18,2004 in Hartnell field, Salinas, CA that had been inoculated with IV-BNYVV from Rockwood 158, Imperial Valley, CA. Scored for reaction to BNYVV on a scale of 0 to 9 where 0 is nondiseased and 9 is dead. DI = disease index, the average score per root. %R = % resistant, where 0 & 1's considered highly resistant.

Table 2. Cultivar response to IV-BNYVV in Coded Trial in 2004 in Rockwood 156, Imperial Valley[a,b,c]

Cultivar		Sugar Yield kgha[-1]	% Sugar	Root Rot %
Rz1 commercial checks				
Beta4430R		6400	8.9	48
Beta4776R		7000	8.7	40
Coded commercial hybrids				
Betaseed	- 1	7700	9.3	40
	- 2	6000	9.9	38
	- 3	14000	11.8	24
	- 4	10600	10.8	41
Spreckels	- 1	5200	9.3	32
	- 4	2300	7.3	66
	- 7	9500	11.1	42
	- 8	6000	9.7	48
USDA-ARS- 2 (*Rz1*)		5700	10.0	50
	- 1 (*Rz1*)	2900	7.5	71
LSD (.05)		3700	1.6	

[a]Commercially harvested fields of Beta4430R at this time were running about 16.7% sugar and 16200 kgha[-1] sugar with very little root rot.

[b]Test was organized and entries coded by the California Beet Growers Association, Stockton, CA. Hybrid cultivars were submitted as coded entries by Betaseed, Holly Hybrids (Spreckels), and USDA-ARS. The specific pedigree and resistance factors were known only to the submitting organization.

[c]Test planted September 2003 into Rockwood 156 where rhizomania occurred in Beta4430R (*Rz1*) in 2002-2003. There were 2-row plots with 8 replications. At harvest, roots were weighed and roots with rot counted by Holly Hybrids. Sub-samples of roots were analyzed for sucrose concentration by Spreckels Sugar Co., Brawley, CA.

INDEX OF VARIETY TRIALS, SALINAS, CA, 2004
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Tests were located in three field plot areas at Salinas and two at Brawley, CA. Disease nurseries were also used in Idaho, Colorado, and Minnesota. Tests at Brawley (Imperial Valley) were planted in September 2003, and harvested from May through June, 2004. Tests at Salinas were planted from March through August, 2004, and harvested from September through December. Tests at Spence Field (Salinas) were under both rhizomania and nonrhizomania (following methyl bromide fumigation) conditions. Herbicides were not used in Block 6 trials that followed strawberries and methyl bromide fumigation. Nortron, Pyramin, Betamix, Progress, and Poast were used in the other trials. Bayleton at 2lbs material/acre was used for powdery mildew control. Lorsban-4E was applied for aphid and other insect control. The specific planting and harvest dates as well as plot size and design are shown on each test summary.

Tests are listed in the main Table of Contents for Salinas by types of material and evaluation. As an aid to find test summaries, they are listed below by ascending test (planting date) number and cross- referenced to the page number. Tests shown as n/a are not available or not included in this report.

TEST NO.	NO. ENTRIES	TEST DESCRIPTION	PAGE NO.

VIRUS YELLOWS, YIELD & PROGENY TESTS, FEBRUARY, 2004 (cont.)

Progeny Tests (cont.)

TEST NO.	NO. ENTRIES	TEST DESCRIPTION	PAGE NO.
1304	96	S_1's from F_1 (MMS^{f}aa x C78/3)	n/a
1404	96	Sn's from selected MMS^{f}Aa lines	n/a
1504	32	Sn's from CR/RZM lines	n/a
1604	48	S_1's from 2943 polycross (%S)	n/a
1704	32	FS's from R276-89	n/a
1804	48	FS's from R_z2 germplams lines	n/a
1904	48	mm lines and populations	A85
2004	48	S_1 & FS's from 812 & 819R_z2 lines	n/a
2104	16	Sn's from C927-4 & 2921	n/a

Powdery Mildew Evaluation Tests

TEST NO.	NO. ENTRIES	TEST DESCRIPTION	PAGE NO.
2204	48	FS's from WB97 & WB242 lines	
2304	32	Powdery mildew resistant lines	A78
2404	48	Coded powdery mildew	A80

SUGARBEET CYST NEMATODE (SBCN)/RHIZOMANIA TESTS, APRIL 2004

Soil infested with SBCN/BNYVV

TEST NO.	NO. ENTRIES	TEST DESCRIPTION	PAGE NO.
3104	96	Progeny performance under SBCN/RZM	n/a
3204	48	Lines under SBCN/RZM	A65
3304	24	Hybrids under SBCN/RZM	A114
3404	12	Mother root selection	n/a
3504	24	Commercial & Exp. hybrids under RZM/SBCN	A116

RHIZOMANIA YIELD, PROGENY SELECTION TRIALS, MAY, 2004

Specialty, Progeny, & Selection Tests

TEST NO.	NO. ENTRIES	TEST DESCRIPTION	PAGE NO.
4104	48	Plant Introductions & Wild Beets	A166
4204	24	mm, RZM progeny test	n/a
4304	12	Root knot nematode lines	A82
4404	48	MM selected progeny lines	A83
4504	96	S_1's from F_1 (MMS^{f}aa x Y91)	n/a
4604	96	S_1's from F_1 (MMS^{f}aa x C78/3)	n/a
4704	96	Sn's from selected MMS^{f}Aa lines	n/a
4804	6	Mother root selection	n/a
4904	48	S_1's from 2943 polycross (%S)	n/a
5004	48	FS's from R_z2 germplasm lines	n/a
5104	32	FS's from R276-89	n/a
5204	48	mm lines and poulations	A85
5304	48	S_1 & FS's from 3812, 3819	n/a

TEST 504. PERFORMANCE OF LINES, SALINAS, CA, 2004

48 entries x 8 reps., RCB(e), 3 subtests: 16 x 8 reps, RCB(e)
1-row plots, 22 ft. long

Planted: March 19, 2004
Harvested: September 27, 2004
BYV Not Inoculated

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/ 100' No.	RJAP %	Powdery Mildew 9/24
504-1: Multigerm, O.P. lines							
Y391H50	C790-15CMS x RZM-ER-% Y191	16079	47.26	17.01	160	85.5	2.3
Phoenix	9/12/03	17193	52.25	16.45	162	88.2	3.8
Beta 4430R	9/21/03	17603	52.96	16.61	159	87.0	2.8
Y369	RZM-ER-% Y169, (C69/8)	14663	42.38	17.35	160	83.9	1.6
03-US75	Inc. 00-US75	10617	38.95	13.64	162	84.1	5.8
03-C37	Inc. U86-37, 99-C37	11727	38.90	15.01	158	83.9	4.9
99-C31/6	Inc. F86-31/6, (C31/6)	13521	43.33	15.59	160	84.9	2.4
R276-89	RZM-% R076-89	13191	40.11	16.46	157	84.0	1.5
R376-89-4	Inc. R176-89-4, (C76-89-4)	11974	36.03	16.64	163	84.1	1.8
R381-22	Inc. R181-22, (C81-22)	14280	42.48	16.79	153	85.1	1.9
Z210	Inc. Z010(C), (Polish %s gp)	13846	37.59	18.38	156	86.1	4.9
99-C46/2	Inc. U86-42/2, (C46/2)	12804	38.70	16.50	157	84.9	3.4
R378	RZM-ER-% R178, (C78/3)	14676	44.91	16.39	156	83.9	1.6
R380	RZM-ER-% R180, (C80/2)	13676	40.66	16.81	162	83.3	1.8
Y390	Inc. Y190-#(C),C2,Syn 1	14778	43.69	16.94	161	84.5	2.6
03-SP22-0	Inc. 01-SP22-0	12036	39.65	15.14	162	84.3	2.3
Mean		13916.5	42.49	16.36	159.3	84.9	2.8
LSD (.05)		1379.4	3.79	0.80	8.7	2.1	1.0
C.V. (%)		10.0	9.00	4.94	5.5	2.5	34.1
F value		15.4**	13.07**	14.94**	0.9NS	3.0**	15.5**

TEST 504. PERFORMANCE OF LINES, SALINAS, CA, 2004
48 entries x 8 reps., RCB(e). ANOVA across tests to compare means.

		Sugar Lbs	Beets Tons	Sucrose %	Beets/ 100' No.	RJAP %	Powdery Mildew 9/24
Mean		14328.7	43.56	16.44	157.3	84.1	2.5
LSD (.05)		1393.1	3.87	0.74	9.1	2.0	0.9
C.V. (%)		9.9	9.01	4.55	5.8	2.4	37.3
F value		7.9**	6.69**	7.91**	2.7**	3.2**	13.2**

TEST 504. PERFORMANCE OF LINES, SALINAS, CA, 2004

(cont.)

| Variety | Description | Acre Yield | | Sucrose | Beets/100' | RJAP | Powdery Mildew |
		Sugar Lbs	Beets Tons	%	No.	%	9/24
504-2:	Multigerm lines with *Bvm* gp						
Y391	RZM-ER-% Y191	14389	42.53	16.92	158	84.8	1.9
Y392	RZM Y292	14896	44.31	16.81	154	83.9	3.5
Y393	Inc. FS-#s,C1,Syn 1	15946	47.26	16.88	155	84.7	1.3
Y375	RZM Y275	15256	45.75	16.67	153	83.7	2.5
R321	RZM R221, (C26 x C27)	15030	46.17	16.27	156	85.5	2.0
Y367	RZM-ER-% Y167, (C67/2)	15578	44.80	17.40	160	84.4	0.9
Y371	RZM-ER-% Y171	14406	43.59	16.54	156	85.2	3.5
R343	RZM-ER-% R143	14590	44.19	16.51	156	83.6	2.5
R336	RZM-ER-% R136, (C79-8)	13401	42.43	15.85	154	82.5	3.4
R340	RZM-ER-% R140	15446	46.71	16.55	168	83.3	1.6
P327	PMR-RZM P227, (CP03)	14048	44.67	15.74	151	83.3	1.8
P328	PMR-RZM P228, (CP04)	15906	49.03	16.23	157	83.3	1.3
P329	PMR-RZM P229, (CP05)	15432	47.67	16.15	156	85.1	0.6
P330	PMR-RZM P230, (CP06)	15391	44.90	17.14	156	84.0	0.9
P318-6	PMR-RZM P118-6, (CP08)	14524	44.79	16.21	154	83.1	0.9
P307/8	PMR-RZM P207/8 (Iso), (CP07)	15080	45.60	16.52	131	82.8	1.1
Mean		14957.5	45.28	16.53	154.6	84.0	1.8
LSD (.05)		1340.0	3.84	0.65	6.9	1.9	0.8
C.V. (%)		9.1	8.58	3.98	4.5	2.3	43.3
F value		2.1*	1.72NS	3.70**	9.0**	1.7NS	12.0**

A55

TEST 504.   PERFORMANCE OF LINES, SALINAS, CA, 2004
(cont.)

Variety	Description	Acre Yield		Sucrose %	Beets/100' No.	RJAP %	Powdery Mildew 9/24
		Sugar Lbs	Beets Tons				
504-3: Multigerm, s^f, Aa populations							
3931	RZM 2931,1931aa x A, (C931)	15164	45.35	16.75	151	82.9	2.6
3941	RZM 2941,1941aa x A, (C941)	15548	48.00	16.21	156	86.5	2.5
3942	RZM 2942aa x A	12801	38.80	16.50	157	82.5	1.8
CR311	RZM CR211, CR111, CR111(C)aa x A, (CR11)	15045	45.95	16.38	158	82.8	3.1
Z325	RZM Z225,Z125(C)aa x A, (Cz25)	14898	43.43	17.16	152	85.0	3.8
3943	2943(C)aa x A	16113	46.51	17.35	159	84.5	2.1
N312	PMR-RZM-NR N112 (A,aa), (CN12)	14845	45.55	16.30	164	82.9	1.0
N372	RZM-ER-NR N172 (A,aa), (CN72)	14803	44.50	16.61	155	83.3	1.8
R324/5	Inc. R824, (C79-2,-3; WB41,42)	13336	40.97	16.34	162	83.1	4.0
R324	Inc. R724, (C79-2; WB41)	12599	40.81	15.46	165	81.9	4.6
R325	Inc. R725, (C79-3; WB42)	12918	40.73	15.86	156	83.2	3.6
R337	Inc. R637, (C79-9; WB151)	13127	40.11	16.35	160	83.3	3.6
P318-6	Inc. P118-6, (CP08)	13452	42.98	15.66	160	83.9	1.8
3849	RZM 2251-2255(C)aa x A	14512	41.32	17.61	156	83.1	3.1
3842	RZM 2842(C)mmaa x A, (C842)	12697	38.19	16.61	159	82.3	4.0
3869	RZM 1869(C)mmaa x A, (C869)	13932	43.64	15.96	159	85.7	3.9
Mean		14112.1	42.93	16.45	158.1	83.6	3.0
LSD (.05)		1347.4	3.98	0.68	8.1	1.9	0.8
C.V. (%)		9.6	9.36	4.15	5.2	2.3	27.4
F value		5.6**	4.19**	5.84**	1.7NS	3.5**	13.6**

NOTES: Tests 104 and 504 are companion tests. Test 104 was inoculated with BYV on May 13, 2004.
Test 504 remained fairly free of yellowing symptoms throughout most of the season. Powdery mildew and
pests were controlled. Except for PM and black aphids, no other disease or pest problems were noted,
except at harvest when rhizomania susceptible entries showed moderate rhizomania.

These tests were grown in Block 6. This field had been in sugarbeet four years earlier. Following
sugarbeet, the field was fumigated with methyl bromide/chloropricin and strawberries grown. These tests
followed the 2003 strawberry crop.

TEST 404. PERFORMANCE OF PROGENY LINES SELECTED FOR VIRUS YELLOWS RESISTANCE WITHOUT INOCULATION, SALINAS, CA, 2004

12 entries x 8 reps., RCB(e)
1-row plots, 22 ft. long

Planted: March 19, 2004
Harvested: October 14, 2004
Not BYV Inoculated

| Variety | Description | Acre Yield | | Sucrose | Beets/100' | RJAP | Powdery Mildew |
		Sugar Lbs	Beets Tons	%	No.	%	9/27
Checks							
03-SP22-0	Inc. 01-SP22-0	13550	43.43	15.60	162	84.7	2.1
R376-89-5-4	RZM R176-89-4-5	14474	44.02	16.45	155	83.7	0.8
Progeny lines from full-sibs							
Y381-22	RZM R181-22, (C81-22)	14870	43.93	16.92	144	84.2	2.6
P318-6	Inc. P118-6, (CP08)	14446	45.40	15.90	152	82.9	1.9
R280/2-9	Inc. R080/2-9	13540	40.55	16.71	149	82.0	4.1
R380-21	RZM R180-21	15232	44.24	17.21	152	83.8	4.4
Y368-8	RZ Y168-8	14268	41.62	17.11	156	84.8	2.6
Y367-5	RZM Y167-5	13777	42.87	16.08	156	84.2	1.6
R376-6	RZM R178-6	14043	43.28	16.23	141	84.1	2.0
Progeny lines from S₁'s							
3931-56	RZM 1931-56aa x A	15609	45.84	17.04	153	83.0	1.3
Z331-14	RZM Z131-14aa x A	15135	42.98	17.60	153	84.7	3.4
2930-19	RZM 1930-19aa x A	15266	44.87	17.04	149	85.3	1.6
Mean		14517.6	43.59	16.66	152.0	84.0	2.4
LSD (.05)		1100.0	3.01	0.52	9.9	1.8	0.9
C.V. (%)		7.6	6.94	3.14	6.5	2.2	36.5
F value		3.3**	1.97*	10.69**	2.5*	2.1*	13.4**

TEST 104. PERFORMANCE OF LINES UNDER BYV INFECTION, SALINAS, CA, 2004

24 entries x 8 reps., RCB(e)
1-row plots, 22 ft. long

Planted: March 19, 2004
Harvested: October 14, 2004

Variety	Description	Sugar Lbs	Beets Tons	Loss %	Sucrose %	Beets/100' No.	RJAP %	Powdery Mildew 9/27	Virus Yellows 7/28	8/06	8/19	Mean
Hybrid checks												
Phoenix	9/12/03	12425	40.35	27.7	15.40	154	86.3	2.5	3.9	4.5	4.9	5.1
Beta 4430R	8/21/03	14183	43.84	19.4	16.16	155	86.0	3.8	5.5	5.1	6.0	6.0
Y391H50	C790-15CMSxRZM-ER-% Y191	13439	42.37	16.4	15.88	159	85.6	1.1	2.0	1.1	2.1	2.6
O.P. MM line checks												
03-SP22-0	Inc. 01-SP22-0	6515	27.41	45.9	11.69	162	83.0	1.3	5.8	5.9	6.5	6.5
03-US75	Inc. 00-US75	6169	28.97	41.9	10.73	154	80.3	4.0	4.5	4.4	6.0	5.1
03-C37	Inc. U86-37, 99-C37	8754	31.90	25.4	13.65	159	82.5	4.4	1.9	1.5	3.8	2.8
99-C46/2	Inc. U86-46/2, (C46/2)	9797	32.71	23.5	14.94	153	84.3	1.4	2.9	1.9	3.5	3.3
99-C31/6	Inc. F86-31/6, (C31/6)	11323	39.20	16.3	14.45	152	85.7	1.5	1.6	0.6	1.5	1.9
MM,O.P. lines												
R276-89	RZM-% R076-89	12365	39.10	6.3	15.81	149	83.3	0.5	1.0	0.3	0.5	1.1
R376-89-4	Inc. R176-89-4	11854	36.15	1.0	16.39	152	82.1	0.8	1.0	0.0	0.3	0.9
R381-22	Inc. R181-22	12485	38.74	12.6	16.08	140	82.7	1.1	2.5	1.6	1.3	2.4
Y369	RZM-ER-% Y169	12465	38.26	15.0	16.30	153	83.3	0.9	2.8	1.6	2.3	2.9
Z210	Inc.Z010(C),(Polish%s *gp*)	9834	31.74	29.0	15.44	155	83.0	4.5	5.5	5.3	6.3	6.0
Y390	Inc.Y190-#(C),C2,Syn 1	11937	37.57	19.2	15.90	152	84.4	1.1	2.1	1.5	3.0	2.8
Y391	RZM-ER-% Y191	12592	39.00	12.5	16.15	157	82.7	0.8	2.3	1.4	2.5	2.8
Y392	RZM Y292	11739	36.53	21.2	16.07	158	83.1	2.5	3.0	3.1	3.4	3.8
R378	RZM-ER-% R178	12184	38.19	17.0	15.93	153	82.1	1.1	3.1	1.6	3.1	3.4
R380	RZM-ER-% R180	12842	39.86	6.1	16.10	157	83.3	0.6	2.9	2.0	2.4	3.2
Y393	Inc. FS-#'s,C1,Syn 1	13561	43.31	15.0	15.66	152	83.6	0.6	1.9	1.1	1.5	2.2
Y375	RZM Y275	12531	38.99	17.9	16.08	156	83.5	0.9	3.3	2.5	2.9	3.3
R321	RZM R221	12347	39.30	17.9	15.70	157	83.8	1.4	2.5	1.5	1.8	2.6

TEST 104. PERFORMANCE OF LINES UNDER BYV INFECTION, SALINAS, CA, 2004

(cont.)

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Loss %	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	RJAP %	Powdery Mildew 9/27	Virus Yellows Scores 7/28	8/06	8/19	Mean
MM,S[f] Aa populations												
3931	RZM 2931, 1931aa x A	13631	10.1	42.79	15.94	156	82.6	1.8	2.8	2.3	3.1	3.2
3941	RZM 2941, 1941aa x A	13324	14.3	41.82	15.94	151	83.7	2.1	2.9	1.8	2.6	3.0
3942	RZM 2942aa x A	12584	1.7	38.50	16.35	152	82.8	1.1	2.5	2.0	2.1	2.8
Mean		11703.4		37.78	15.36	154.1	83.5	1.7	2.9	2.3	3.1	3.3
LSD (.05)		1246.6		3.15	1.07	8.9	2.6	0.8	0.8	0.7	0.9	0.5
C.V. (%)		10.8		8.46	7.08	5.9	3.2	47.6	26.9	31.7	30.3	15.6
F value		21.5**		14.91**	13.87**	1.8*	2.1**	17.9**	21.9**	39.8**	29.0**	63.3**

NOTES: Tests 104 and 504 are companion tests. Test 104 was inoculated May 13, 2004 with *Beet yellows virus* (BYV). %Loss is the relative sugar yield loss calculated from the corresponding means in each test. Inoculum was produced by H.-Y.Liu and J.L.Sears. A source of BYV was passed through *chenopodium captitatum* and from plants with severe vein clearing, transferred to sugarbeet plants used to produce viruliferous aphids for the field inoculation. BWYV and BChV could not be detected in the source plants or subsequently from plants inoculated in the field. Little natural VY infection appeared to occur.

Virus yellows foliar symptoms were scored on a scale of 0-9, where 9 = 90-100% of the mature leaf area yellowed. Scores were made on 7/28, 8/06, 8/19 by DP.

At harvest, test 104 showed moderate rhizomania in susceptible entries, e.g., 03-SP22-0, 03-US75, 03-C37, 99-C46/2, 99-C31/6, and Z210.

Correlations within VY inoculated test 104

	SY	RY	%S	RJAP	% loss
BYV mean	-0.43*	-0.39NS	-0.46*	0.07NS	0.77**
BYV 7/28	-0.40NS	-0.37NS	-0.42*	0.02NS	0.71**
BYV 8/06	-0.44**	-0.40NS	-0.46*	0.05NS	0.74**
BYV 8/19	-0.55**	-0.50*	-0.57**	0.00NS	0.82**
% sugar	0.90**	0.75**		0.33NS	-0.82**
% loss	-0.78**	-0.68**	-0.82**	-0.06NS	

Correlations between corresponding tests

	Non-inoculated test (Test 504) SY	RY	%S	RJAP
VY Inoc.				
SY	0.77**	0.59**	0.64**	0.14NS
RY	0.79**	0.71**	0.46*	0.20NS
% sugar	0.60**	0.31NS	0.82**	0.04NS
RJAP	0.69**	0.66**	0.31NS	0.63**
% loss	-0.20NS	0.00NS	-0.49*	0.30NS

TEST 304. PERFORMANCE OF PROGENY LINES SELECTED FOR VIRUS YELLOWS RESISTANCE, SALINAS, CA, 2004

Planted: March 19, 2004
Harvested: October 14, 2004
Inoculated BYV: May 13, 2004

12 entries x 8 reps., RCB(e)
1-row plots 22 ft. long

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Loss %	Acre Yield Beets Tons	Sucrose %	Beets/ 100' No.	RJAP %	Powdery Mildew 9/27	Virus Yellows Scores 7/28	8/06	8/19	Mean
Checks												
03-SP22-0	Inc. 01-SP22-0	9263	31.6	31.64	14.63	158	82.1	1.0	6.6	6.9	7.0	7.1
R376-89-5-4	RZM R176-89-4-5	12338	14.8	38.19	16.13	151	83.7	0.1	1.6	1.0	1.0	1.8
Progeny lines from full-sibs												
Y381-22	RZM R181-22, (C81-22)	12695	14.6	38.85	16.38	155	83.4	1.5	2.9	2.0	1.8	2.9
P318-6	Inc. P118-6, (CP08)	12426	14.0	41.02	15.14	154	83.5	1.0	1.9	1.5	1.8	2.4
R280/2-9	Inc. R080/2-9	11635	14.1	35.17	16.54	141	82.1	3.0	4.4	4.1	5.3	5.1
R380-21	RZM R180-21	12086	20.7	37.32	16.20	153	82.6	2.8	5.1	4.8	5.1	5.3
Y368-8	RZ Y168-8	11944	16.3	35.93	16.66	159	84.7	2.8	3.8	3.4	3.0	3.7
Y367-5	RZM Y167-5	11983	13.0	36.83	16.29	148	82.8	0.9	3.5	4.3	4.3	4.3
R376-6	RZM R178-6	10426	25.8	33.15	15.73	143	84.5	1.8	5.5	5.4	6.1	6.0
Progeny lines from S1's												
3931-56	RZM 1931-56aa x A	12401	20.6	37.98	16.31	150	83.4	1.0	2.3	0.9	0.5	1.9
2331-14	RZM Z131-14aa x A	13964	7.7	39.99	17.45	144	83.3	2.3	3.0	2.9	1.6	3.0
2930-19	RZM 1930-19aa x A	12234	19.9	38.30	15.98	148	82.9	0.4	2.1	1.3	1.3	2.4
Mean		11950.0		37.03	16.12	150.4	83.3	1.5	3.6	3.2	3.2	3.8
LSD (.05)		1127.8		3.08	0.76	10.8	2.1	1.0	0.7	1.0	1.0	0.6
C.V. (%)		9.5		8.34	4.76	7.2	2.5	64.5	20.6	30.1	31.7	15.9
F value		8.5**		6.15**	7.11**	2.1*	1.3NS	7.7**	37.2**	32.4**	38.1**	64.3**

NOTES: Tests 304 and 404 are companion tests. Test 304 was inoculated May 13, 2004 with *Beet yellows virus* (BYV). %Loss is the relative sugar yield loss calculated from the corresponding means in each test. Inoculum was produced by H.-Y.Liu and J.L.Sears. A source of BYV was passed through *chenopodium captitatum* and from plants with severe vein clearing, transferred to sugarbeet plants used to produce viruliferous aphids for the

(cont.)

Variety	Description	Acre Yield			Sucrose	Beets/100'	Powdery Mildew RJAP 9/27	Virus Yellows Scores			
		Sugar Lbs	Loss %	Beets Tons	%	No.	%	7/28	8/06	8/19	Mean

NOTES (cont.):

field inoculation. BWYV and BChV could not be detected in the source plants or subsequently from plants inoculated in the field. Little natural VY infection appeared to occur.

Virus yellows foliar symptoms were scored on a scale of 0-9, where 9 = 90-100% of the mature leaf area yellowed. Scores were made on 7/28, 8/06, 8/19 by DP.

At harvest, tests 304 and 404 showed moderate rhizomania.

Correlations within VY inoculated test 304

	SY	RY	%S	RJAP	% loss
BYV mean	-0.78**	-0.85**	-0.37NS	-0.32NS	0.61*
BYV 7/28	-0.76***	-0.86**	-0.33NS	-0.27NS	0.65*
BYV 8/06	-0.73**	-0.82**	-0.31NS	-0.29NS	0.54NS
BYV 8/19	-0.79***	-0.84**	-0.40NS	-0.32NS	0.58NS
% sugar	0.75***	0.40NS		0.22NS	-0.73**
% loss	-0.88***	-0.76**	-0.73*	-0.14NS	

Correlations between corresponding tests

	Non-inoculated test (Test 404)			
VY Inoc.	SY	RY	%S	RJAP
SY	0.65*	0.18NS	0.71**	-0.05NS
RY	0.65*	0.46NS	0.44NS	-0.11NS
% sugar	0.37NS	-0.37NS	0.84**	0.03NS
RJAP	0.18NS	0.06NS	0.18NS	0.23NS
% loss	-0.20NS	0.20NS	-0.46NS	0.19NS
BYV mean	-0.63*	-0.48NS	-0.40NS	0.07NS

TEST 5404. PERFORMANCE OF LINES UNDER RHIZOMANIA, SALINAS, CA, 2004

48 entries x 8 reps., RCB(e), 3 subsets: 16x8, RCB(e)
1-row plots, 22 ft. long

Planted: May 4, 2004
Harvested: October 14, 2004

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/ 100' No.	RJAP %	Bolting %
5404-1: Multigerm, O.P. lines							
Y391H50	C790-15CMS x RZM-ER-% Y191	11428	34.47	16.58	198	82.4	-.-
Phoenix	9/12/03	11730	38.19	15.36	206	82.4	-.-
Beta 4430R	9/21/03	12453	38.84	16.02	200	83.3	-.-
Y369	RZM-ER-% Y169, (C69/2)	11684	34.92	16.73	205	82.8	-.-
03-US75	Inc. 00-US75, susc.check	9067	32.25	14.05	197	79.2	-.-
03-C37	Inc. U86-37, 99-C37, susc.check	8888	28.80	15.41	214	82.6	-.-
99-C31/6	Inc. F86-31/6, (C31/6), susc.check	9354	31.24	14.94	190	83.4	-.-
R276-89	RZM-% R076-89	9649	30.18	15.96	198	81.3	-.-
R376-89-4	Inc. R176-89-4,(C76-89-4)	9343	28.17	16.58	197	83.7	-.-
R381-22	Rzm R181-22,(C81-22)	10707	32.84	16.30	184	82.2	-.-
Z210	Inc. Z010(C), (Polish %S gp)	10324	29.63	17.44	189	81.2	-.-
99-C46/2	Inc. U86-42/2, (C46/2), susc.check	9180	29.50	15.55	165	82.1	-.-
R378	RZM-ER-% R178,(C78/3)	11385	34.77	16.39	201	82.1	-.-
R380	RZM-ER-% R180,(C80/2)	11018	33.61	16.39	199	82.5	-.-
Y390	Inc. Y190-#(C),C2,Syn 1	11612	35.02	16.59	198	82.7	-.-
03-SP22-0	Inc. 01-SP22-0, susc.check	8262	28.22	14.60	200	83.5	-.-
Mean		10380.4	32.54	15.93	196.3	82.3	-.-
LSD (.05)		716.8	1.98	0.50	12.6	2.0	-.-
C.V. (%)		7.0	6.16	3.14	6.5	2.4	-.-
F value		24.9**	22.59**	25.05**	5.9**	2.4*	-.-

TEST 5404. PERFORMANCE OF LINES UNDER RHIZOMANIA, SALINAS, CA, 2004
48 entries x 8 reps., RCB(e). ANOVA across tests to compare means.

		Sugar Lbs	Beets Tons	Sucrose %	Beets/100' No.	RJAP %	Bolting %
Mean		10626.3	33.24	15.98	196.5	81.9	0.1
LSD (.05)		721.1	1.94	0.58	13.1	2.3	0.4
C.V. (%)		6.9	5.93	3.69	6.8	2.9	271.4
F value		14.1**	14.87**	10.46**	3.0**	2.0**	52.0**

TEST 5404. PERFORMANCE OF LINES UNDER RHIZOMANIA, SALINAS, CA, 2004

(cont.)

| Variety | Description | Acre Yield | | Sucrose | Beets/ 100' | RJAP | Bolting |
		Sugar Lbs	Beets Tons	%	No.	%	%
5404-2: Multigerm lines with *Bvm* gp							
Y391	RZM-ER-% Y191	10295	31.19	16.52	191	81.6	0.0
Y392	RZM Y292	10913	33.26	16.40	194	82.5	0.0
Y393	Inc. FS-#s,C1,Syn 1	12065	36.78	16.40	197	83.7	0.0
Y375	RZM Y275	10723	33.31	16.09	202	82.1	0.0
R321	RZM R221, (C26 x C27)	10729	34.11	15.68	198	82.7	0.0
Y367	RZM-ER-% Y167, (C67/2)	10876	32.95	16.50	195	81.7	0.0
Y371	RZM-ER-% Y171	10654	33.41	15.91	204	81.3	0.0
R343	RZM-ER-% R143	11072	34.11	16.24	200	82.8	0.0
R336	RZM-ER-% R136, (C79/8)	10143	33.66	15.04	201	81.2	0.0
R340	RZM-ER-% R140	11259	35.78	15.70	199	80.4	0.0
P327	PMR-RZM P227, (CP03)	10203	33.15	15.39	194	82.1	7.0
P328	PMR-RZM P228, (CP04)	11199	37.34	14.99	199	77.9	0.0
P329	PMR-RZM P229, (CP05)	11229	34.63	16.21	190	82.5	0.0
P330	PMR-RZM P230, (CP06)	11636	35.52	16.36	199	82.0	0.0
P318-6	Inc. P118-6, (CP08)	10619	33.46	15.85	192	81.0	0.0
P207/8(Sp)	Inc. P007/8, (CP07)	10642	33.15	16.06	192	82.4	0.0
Mean		10891.1	34.11	15.96	196.6	81.7	0.4
LSD (.05)		640.8	1.93	0.53	11.1	2.3	0.7
C.V. (%)		5.9	5.71	3.38	5.7	2.9	157.1
F value		5.0*	5.17**	6.62**	1.1NS	2.4*	52.0**

TEST 5404. PERFORMANCE OF LINES UNDER RHIZOMANIA, SALINAS, CA, 2004
(cont.)

| Variety | Description | Acre Yield | | Sucrose | Beets/100' | RJAP | Bolting |
		Sugar Lbs	Beets Tons	%	No.	%	%
5404-3: Multigerm, S^f,Aa populations							
3931	RZM 2931,1931aa x A, (C97)	11367	34.47	16.48	193	82.6	-.-
3941	RZM 2941,1941aa x A, (C94)	11613	37.14	15.64	189	82.4	-.-
3942	RZM 2942aa x A	10230	30.23	16.95	196	82.3	-.-
CR311	RZM CR211,CR111(C)aa x A, (CR11)	11045	35.55	15.56	183	81.4	-.-
Z325	RZM Z225,Z125(C)aa x A, (C25/2)	11433	34.67	16.49	185	82.4	-.-
3943	2943(C)aa x A	11629	34.67	16.76	201	80.8	-.-
N312	PMR-RZM-NR N112 (A,aa), (C12)	10151	31.59	16.11	199	82.9	-.-
N372	RZM-ER-NR N172 (A,aa), (C72)	10686	34.57	15.38	194	79.6	-.-
R324/5	Inc. R824, (C79-2,-3; WB41,42)	10245	32.20	15.91	210	81.0	-.-
R324	Inc. R724, (C79-2; WB41)	9445	31.19	15.11	210	81.2	-.-
R325	Inc. R725, (C79-3; WB42)	8978	28.28	15.88	202	80.1	-.-
R337	Inc. R637, (C79-9; WB151)	9996	30.59	16.34	210	81.9	-.-
P318-6H5(Sp)	C833-5HO x P118-6, (CP08)	12382	37.53	16.46	186	80.3	-.-
3849	RZM 2251-2255(C)aa x A	11209	33.46	16.75	194	81.9	-.-
3842	RZM 2842(C)mmaa x A, (C84)	9335	29.12	16.04	194	82.6	-.-
3869	RZM 1869(C)mmaa x A, (869)	9974	33.76	14.75	198	80.6	-.-
Mean		10607.4	33.06	16.04	196.6	81.5	-.-
LSD (.05)		757.3	1.91	0.69	11.7	2.6	-.-
C.V. (%)		7.2	5.83	4.32	6.0	3.2	-.-
F value		12.7**	16.03**	6.56**	4.3**	1.2NS	-.-

NOTES: See Test 104 for lines under Beet yellows virus conditions and 504 for Nondiseased performance.
Rhizomania yield tests were run in Block 2 at Spence field. This area in the 1990s was one of the best
locations for rzm tests, but became severely infected with *Sclerotium rolfsii*. Four years ago, it was
fumigated with methyl bromide to eliminate *Sclerotium*. In 2002, strawberries were grown. In 2003, except
for winter oat cover crops, it remained fallow until August, when it was prepared for future rzm tests.
Rhizomania soil was broadcast over the area and disked in. A susceptible sugarbeet variety was then
drilled in and grown for 4 months, then disked in. In April 2004, rhizomania tests were planted. However,
it is now apparent that the level of rhizomania was low at planting time and at worst was only mild through
the course of the season.

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TEST 3204. PERFORMANCE OF LINES WITH RESISTANCE TO RHIZOMANIA AND SBCN, SALINAS, CA, 2004, BLOCK 4

48 entries x 8 reps., sequential
1-row plots, 11 ft. long

Planted: April 26, 2004
Harvested: October 27, 2004

Variety	Resistance	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	Beets/100' No.	RJAP %	Bolting %
Checks										
HH 142	Rz	9-12-03	10271	30.53	16.88	16	15	148	80.2	0.0
Beta 4430R	Rz	8-21-03	10940	32.06	17.14	17	17	157	82.9	0.0
Angelina	Rz1,Rz2		11511	34.58	16.71	18	17	164	79.1	0.0
Roberta	--		6769	23.09	14.68	18	17	169	81.1	0.0
US H11	--	1999 production	6688	22.48	14.95	18	17	170	79.8	0.0
1927-4H5	Rz,R22	C833-5CMS x RZM 9929-4,(C927-4)	10792	32.00	16.90	17	18	162	80.5	0.0
R378H93	Rz,Bp	N265-31HO x RZM R178, (C78/3)	8919	27.36	16.33	16	15	152	78.7	0.0
P318-6H5	Rz,WB242	2833-5HO x P118-6,(CP08)	10100	29.98	16.91	18	18	168	79.0	0.0
Multigerm breeding lines										
R378(Iso)	Rz	RZM-ER-% R178, (C78/3)	10358	30.32	17.11	18	18	170	80.3	0.0
Y390	Rz	Inc. Y190-#(C),C2,Syn1	9430	28.04	16.85	19	18	178	79.9	0.0
Y391	Rz,R22	RZM-ER-% Y191,C2,Syn1	9645	28.70	16.90	16	15	152	78.9	0.0
Y392	Rz,R22	RZM Y292,C1,Syn2	10570	32.30	16.39	18	17	164	80.0	0.0
Y393	Rz,R22	RZM FS-#(C),C1,Syn1	10611	32.71	16.24	17	16	156	79.9	0.0
Y375	Rz,R22	RZM Y275	10090	30.98	16.31	17	16	155	79.9	0.0
Y321	Rz,Bvm	RZM R221, (C26,C27)	11113	34.80	16.00	18	16	164	80.1	0.0
Y367	Rz,R22	RZM-ER-% Y167,(C67/2)	9355	27.41	17.10	17	16	155	80.2	0.0
Y371	Rz,R22	RZM-ER-% Y171	10498	31.63	16.65	18	17	166	79.7	0.0
03-C37	--	Inc. U86-C37	6276	19.30	16.24	17	17	162	79.0	0.0
P328	Rz,WB242	PMR-RZM P228,(CP04)	9587	30.87	15.56	18	17	168	79.4	0.0
P327	Rz,WB97	PMR-RZM P227,(CP03)	7831	24.98	15.66	18	18	169	78.3	5.2
R336	R22	RZM-ER-% R136,(C79-8)	8943	29.10	15.38	19	18	180	77.8	0.0
R340	R22	RZM-ER-% R140	10966	32.91	16.67	18	18	172	80.4	0.0
R324/5	WB41/42	Inc. R824, (C79-2,-3)	6837	20.86	16.40	19	18	176	78.9	0.0
R337	WB151	Inc. R637,(C79-9,WB151)	7605	22.98	16.60	20	18	183	78.1	0.0

TEST 3204.   PERFORMANCE OF LINES WITH RESISTANCE TO RHIZOMANIA AND SBCN, SALINAS, CA, 2004, BLOCK 4

(cont.)

Variety	Resistance	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	Beets/100' No.	RJAP %	Bolting %
Multigerm breeding lines (cont.)										
R381-22	Rz	RZM R181-22,(C81-22)	9715	29.25	17.01	17	17	162	81.0	0.0
R343	R22	RZM-ER-% R143	10585	33.00	16.01	17	16	157	79.9	0.0
R243-14	R22	Inc. R043-14	10056	31.54	15.96	16	16	151	79.4	0.0
3927-4	R22,Rz	RZM 2927-4(A,aa),(C927-4)	7694	24.72	15.66	16	16	153	80.3	0.0
N372	Rz,Bvm	RZM-ER-NR N172,(CN72)	9801	30.34	16.21	17	16	159	79.8	0.0
N312	Rz,WB242	PMR-RZM-NR N112,(CN12)	8974	27.74	16.20	17	15	155	78.4	0.0
P329	Rz,WB97	PMR-RZM P229,(CP05)	9909	29.82	16.65	16	16	150	80.2	0.0
P330	Rz,WB242	PMR-RZM P230,(CP06)	9881	30.50	16.23	17	18	159	79.4	0.0
R378(Iso)	Rz	RZM-ER-% R178, (C78/3)	10453	30.84	16.96	18	18	168	80.0	0.0
P207/8(Sp)	Rz,WB97/242	Inc. P007/8,(CP07)	8856	26.95	16.54	18	17	165	79.1	0.0
P207/8H5	Rz,WB97/242	C833-5HO x P007/8,(CP07)	10696	32.01	16.73	16	15	150	80.3	0.0
N172	Rz,Bvm	NR-RZM N972 (A,aa)	8208	26.39	15.49	17	15	159	80.6	0.0
N324	Rz,Bp	RZM N224 (g)	8789	28.93	15.24	16	16	152	79.3	0.0
N325	Rz,Bp	RZM N224(C) (g)	6821	20.84	16.39	17	15	157	78.8	0.0
P318-6(Iso)	Rz,WB97/242	PMR-RZM P118-6,(CP08)	8444	26.45	15.95	18	17	165	78.6	0.0
P318-6(Sp)	Rz,WB97/242	Inc. P118-6,(CP08)	8486	26.56	16.08	18	17	170	78.2	0.0
2927-4	R22,Rz	RZM 1927-4,(C927-4)	7310	22.78	16.08	19	17	173	80.1	0.0
Y267-21	R22,Rz	Inc. Y067-21	10004	30.89	16.23	18	17	165	79.7	0.0
Y271-14	R22,Rz	Inc. Y071-14	9659	29.03	16.66	16	15	151	79.5	0.0
Y367-5	R22,Rz	RZM Y167-5	8096	25.95	15.63	17	16	160	80.0	0.0
Y375-9	Rz,R22	Inc. Y175-9	9348	29.75	15.74	17	16	162	79.9	0.0
Y375-13	Rz,R22	Inc. Y175-13	10091	31.53	16.08	16	14	150	79.9	0.0
Y375-20	Rz,R22	Inc. Y175-20	9892	30.42	16.27	17	15	156	79.8	0.0
02-WB242	WB242	Inc. WB242,(C37 x WB242)	3419	11.94	14.34	17	16	158	77.1	0.0

(cont.)

Variety	Resistance	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	Beets/ 100' No.	RJAP %	Bolting %
Mean			9185.3	28.29	16.23	17.8	16.9	161.8	79.6	1.0
LSD (.05)			1075.5	3.48	0.59	1.9	2.1	17.4	2.3	2.1
C.V. (%)			11.9	12.50	3.69	10.9	12.8	10.9	2.9	224.3
F value			16.7**	12.47**	9.10**	1.9**	2.1**	1.9**	1.4NS	60.8**

NOTES: Test 3204 was hand harvested under very wet conditions but not scored for rhizomania. Rhizomania was moderate to severe. Test 3205 was adjacent to Test 3504 and had low to moderate sugarbeet cyst nematode infestation. Powdery mildew was controlled.

Resistance: $Rz = Rz1$ or $Rz2$ or unknown from B.vulgaris subsp. maritima. Bp = cyst nematode resistance from B.procumbens, Hs^{pro-1}. $R22$ = resistance to rhizomania and/or SBCN from C50/C51 germplasm.   WB242 = resistance to powdery mildew (Pm) and possibly SBCN from WB242 accession.   WB97 = resistance to powdery mildew from WB97.   WB41/42 & WB151 = segregation for $Rz2$ at low frequency in C37 background. Bvm = unknown B.vulgaris subsp. maritima source.   The letter "C" has multiple meanings: (C) = composite of progeny lines for re-synthesis; C1 = cycle one from selection, e.g., C2,syn1 means cycle 2 of recurrent selection with seed from first synthesis; C# = released from California (Salinas).

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TEST 6204. EVALUATION OF LINES UNDER RHIZOMANIA AND CERCOSPORA LEAF SPOT, SALINAS, CA, 2004

Planted: May 5, 2004
Harvested: November 16, 2004
Inoc. CLS: August 19, 2004

48 entries x 8 reps., sequential
1-row plots, 11 ft. long

Variety	Description	Acre Yield			Beets/ 100' No.	RJAP %	Cercospora Leaf Spot Score
		Sugar Lbs	Beets Tons	Sucrose %			
Line checks							
Roberta	2/25/04	13602	41.63	16.33	182	85.1	4.9
01-EL0204	RZM OO-EL0204, smooth root sel.	11759	36.20	16.23	167	81.6	2.5
R378	RZM-ER-% R178, (C78/3)	12874	37.69	17.06	193	79.9	2.3
Y390	Inc. Y190-#(C)	13722	38.64	17.73	174	81.6	2.5
Y391	RZM-ER-% Y191	13023	36.38	17.90	191	82.1	2.0
Y392	RZM Y292	12284	35.46	17.29	176	80.0	2.1
Y393	FS#(C)	13330	38.20	17.45	169	81.8	3.0
03-SP22-O	Inc. 01-SP22-0, (SP7622-0)	9728	30.24	16.08	184	82.8	0.6
Z325	RZM Z225,Z125(C)aa x A, (CZ25/2)	13344	37.69	17.75	182	81.5	2.3
Z210	PX %S Polish(C)	12085	30.48	19.84	180	82.1	4.6
Y375	RZM Y275	12525	37.09	16.88	180	81.5	2.3
R321	RZM R221, (C26 x C27)	12411	36.49	17.00	173	82.1	2.0
Lines							
01-FC1030	RZM FC1030aa x A	11148	32.35	17.15	178	81.3	2.3
03-FC1030-15	Inc. 01-FC1030-15 (A,aa)	10217	29.53	17.34	169	77.4	3.0
03-FC1030-16	Inc. 01-FC1030-16 (A,aa)	12254	34.17	17.92	180	81.7	3.1
2933-14	Inc. 0933-14 (A,aa)	10780	30.13	17.90	169	82.7	3.8
3933-107	Inc. 1933-107 (A,aa)	10675	31.21	17.08	169	80.5	3.8
3933-113	Inc. 1933-113 (A,aa)	12503	36.19	17.26	178	81.0	1.9
3933-118	Inc. 1933-118 (A,aa)	11982	33.43	17.91	184	81.5	2.8
3931	RZM 2931,1931aa x A, (C931)	15249	42.73	17.84	183	81.4	2.6
CR311	RZM CR211,CR111(C)aa x A, (CR11)	13060	38.20	17.04	183	81.1	1.6
CR311-6	Inc. CR111-6 (A,aa)	11587	34.99	16.54	186	78.5	1.1
CR311-41	Inc. CR111-41 (A,aa)	11199	33.76	16.58	172	80.6	1.0
CR311-88	Inc. CR111-88 (A,aa)	13806	41.42	16.66	193	80.8	0.9

(cont.)

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	RJAP %	Cercospora Leaf Spot Score
Lines (cont.)							
CRO09-1	RZM CR909-1aa x A, (CR09-1)	9484	28.05	16.95	168	78.2	1.9
CR211-7	Inc. CR911-7 (Sp)	12124	35.80	16.96	177	80.2	1.3
CR110-14-2	Inc. CR910-14-2 (Inc. S_2 line)	8729	26.18	16.70	193	79.8	0.1
CR310-14-2	RZM CR110-14-2 (A,aa)	8280	24.98	16.57	185	80.0	0.0
FC20031018	F_3(9931 x FC709-2), 4/14/04	10290	29.41	17.46	195	79.9	1.9
FC20031019	F_3(FC712 x 9931), 4/14/04	12371	37.92	16.33	189	80.6	2.5
FC20031022	(9931 x FC907) x FC709-2, 4/14/04	10530	28.52	18.42	185	80.9	3.6
CR112-5	Inc. CR812-5, (Inc. S_1 line)	10203	31.14	16.38	202	79.7	2.3
Monogerm lines and populations							
3842	RZM 2842mmaa x A, (C842)	10903	32.17	16.98	199	79.9	2.6
03-FC124	RZM 02-FC124mmaa x A	12344	35.76	17.25	211	80.0	2.5
03-FC1015	RZM 02-FC1015mmaa x A	11361	32.97	17.24	184	80.3	1.8
03-FC123-31	Inc. 01-FC123-31 (A,aa)	10001	29.03	17.17	206	81.0	2.5
03-FC1014-22	Inc. 01-FC1014-22 (A,aa)	10023	27.92	17.95	203	81.8	2.4
03-SP22-O	Inc. 01-SP22-O, (SP7622-0)	8287	27.21	15.07	184	82.2	0.6
Hybrids							
Angelina	2/25/04	11839	35.17	16.83	205	81.6	4.0
Beta 4430R	8/21/03	14821	43.54	17.05	200	82.5	5.0
Monohikari		11939	33.26	17.94	198	83.9	2.8
ACH555	CLSR check, lot# 8107307, 3/8/02	12279	34.32	17.91	178	81.4	1.5
HM-E17	rec'd 3/21/02	12029	33.56	17.90	202	82.5	1.8
Roberta	susc. check, 2/25/04	13347	41.32	16.13	183	86.0	4.1
R378H73	02-FC124HO x R178	13201	37.99	17.41	187	81.3	2.3
R378H74	02-FC1015HO x R178	13410	38.30	17.48	192	81.2	2.0
CR311H5	C833-5HO x RZM CR211	13606	39.21	17.35	187	80.9	1.4
HH142	9/12/03	12698	36.38	17.48	183	82.8	2.3

TEST 6204. EVALUATION OF LINES UNDER RHIZOMANIA AND CERCOSPORA LEAF SPOT, SALINAS, CA, 2004

(cont.)

| Variety | Description | Acre Yield | | Sucrose | Beets/ 100' | RJAP | Cercospora Leaf Spot |
		Sugar Lbs	Beets Tons	%	No.	%	Score
Mean		11859.3	34.47	17.20	185.3	81.2	2.3
LSD (.05)		1251.8	3.45	0.61	19.2	2.4	1.0
C.V. (%)		10.7	10.18	3.63	10.5	3.0	44.6
F value		12.4**	13.20**	11.53**	2.6**	3.2**	9.4**

NOTES: Rhizomania was mild. Test area was inoculated with Cercospora beticola on August 19, 2004 and scored on approximately the KWS scale, just before harvest, where 0 = no disease to 9 = severe disease. CLS development was slow and mild, partially due to a cooler and drier September-October than normal. Extreme reactions were easily seen, but the middle was difficult to separate.

Powdery mildew was controlled until late and then developed on the most susceptible varieties. No other diseases or pests appeared to be significant.

See Test 104 and 504 for performance of lines under virus yellows and nondiseased conditions and Test 5404 for mild rhizomania conditions.

In 2004, CR311 is being released as CR11. The progeny lines CR311-6, -41, -88; CR211-7; CR310-14-2; and CR112-5 were isolated from earlier versions of population CR311 as half-sib and selfed progeny. Monogerm line 03-FC123-31 is a selection from FC301 released in 2004. 03-FC1014-22 is a selection from FC201 released in 2004. FC1030-15 & -16 are selections from FC1030, a population hybrid between Rhizoctonia and rhizomania/CTR germplasm.

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TEST 7104. HARTNELL FIELD EVALUATION OF GERMPLASM FOR REACTION TO IMPERIAL STRAIN OF BNYVV, SALINAS, CA, HARTNELL, 2004

96 entries x 4 reps., sequential
1-row plots, 10 ft. long

Planted: May 5, 2004
Harvested: November 18, 2004

Variety	Description	Sugar Yield Sugar Lbs	Sugar Yield Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	RJAP %	Bolting %	DI	%R(0-1)	%R(0-4)	%R(0-5)
Hybrids												
US H11	susc. ck., 10/14/02	4921	18.79	12.90	24.5	20.8	79.4	0.0	5.0	4.3	31.6	58.0
Rizor	3/30/01	6488	21.47	15.10	24.5	22.0	81.6	0.0	4.3	11.5	52.7	70.1
Beta 4430R	9/21/03	9423	34.08	13.73	25.0	23.0	83.2	0.0	2.4	45.6	88.1	93.5
Angelina	resist. ck., 2/25/04	9462	31.98	14.80	24.0	22.8	81.2	0.0	2.2	54.2	87.6	92.8
HH142	resist. ck., 9/12/03	5450	21.36	12.55	22.3	19.8	82.0	0.0	4.4	14.1	46.3	65.6
Beta 4001R	resist. ck., 8/25/03	8622	28.82	14.88	25.8	21.5	80.8	0.0	3.6	11.7	68.7	82.9
Phoenix	9/12/03	6889	26.44	12.90	21.8	17.0	84.4	0.0	3.7	24.6	60.2	79.5
Roberta	susc. ck., 2/25/04	5828	21.47	13.57	22.5	18.0	84.1	0.0	5.0	4.4	42.2	61.2
R378H47	2848H5 x R178 (C78/3)	7748	26.37	14.63	23.0	22.0	81.7	0.0	3.6	22.2	66.8	79.5
R378H68	2848-1H5 x R178	7038	24.39	14.52	23.3	20.5	80.5	0.0	2.7	36.5	82.9	93.1
R378H80	2810-17H5 x R178	7313	24.62	14.88	25.0	22.0	82.0	0.0	3.4	21.4	66.8	84.9
R378H81	2810-19H5 x R178	6199	21.71	14.40	23.3	18.5	80.5	0.0	3.8	20.5	63.4	78.1
Beta 4430R	9/21/03	7848	29.99	13.43	25.3	23.0	82.4	0.0	3.1	44.2	64.0	75.8
Angelina	resist. ck., 2/25/04	10112	35.24	14.25	23.0	21.0	82.5	0.0	2.3	48.7	84.0	90.9
Y375H50	C790-15CMS x RZM Y275	5273	19.37	13.77	24.8	21.0	81.6	0.0	3.7	22.8	56.5	78.4
Y277H5	C833-5HO x RZM Y175-R136	6464	21.82	14.73	21.8	20.3	82.1	0.0	3.5	22.9	69.2	82.5
Lines with $Rz1$ resistance												
02-US22/3	Inc. 97-US22/3, susc.ck.	4561	15.99	14.27	23.8	19.0	79.1	0.0	4.8	3.2	37.7	60.3
03-US75	Inc. 00-US75, susc.ck.	4457	18.09	12.35	25.0	22.0	76.5	0.0	4.3	8.5	52.7	74.2
03-SP22-0	Inc. 01-SP22-0, susc.ck.	2998	11.79	12.60	24.0	18.8	80.2	0.0	5.1	2.8	34.1	58.9
01-EL0204	RZM 00-EL0204, (EL0204)	7104	27.07	13.07	23.8	21.8	82.3	0.0	2.5	41.6	87.5	95.6
01-FC1030	RZM FC composite aa x A	5159	17.97	14.27	20.0	17.3	80.9	0.0	4.1	10.0	57.8	79.4
Y369	RZM-ER-% Y169, (C69/2)	8616	27.42	15.65	22.5	21.3	82.9	0.0	3.4	19.6	69.1	83.7

TEST 7104. HARTNELL FIELD EVALUATION OF GERMPLASM FOR REACTION TO IMPERIAL STRAIN OF BNYVV, SALINAS, CA, HARTNELL, 2004

(cont.)

		Sugar Yield			Stand Count	Harv Count				Rhizomania Resistance		
Variety	Description	Sugar Lbs	Beets Tons	Sucrose %	No.	No.	RJAP %	Bolting %	DI	%R(0-1)	%R(0-4)	%R(0-5)
Lines with R$_{z1}$ resistance (cont.)												
R380	RZM-ER-% R180, (C80/2)	6913	23.46	14.75	26.5	22.5	80.0	0.0	3.1	28.8	73.6	86.8
R370	RZM-ER-% R170	7170	26.26	13.73	25.8	22.0	80.4	0.0	3.3	24.0	66.0	86.5
Y390	Inc. Y190-#(C)	6790	21.59	15.70	23.0	20.8	83.5	0.0	3.8	17.1	59.0	77.9
Y391	RZM-ER-% Y191	7153	23.92	14.95	24.8	24.5	81.1	0.0	3.4	25.2	72.5	85.9
Y392	RZM Y292	6040	21.36	14.15	22.8	21.3	79.6	0.0	3.2	32.3	67.5	83.7
Y393	Inc. FC-#'s, C1, Syn 1	7397	25.67	14.40	24.3	21.8	84.0	0.0	2.8	34.1	81.6	85.0
R378	RZM-ER-% R178, (C78/3)	6370	20.77	15.43	24.8	22.5	83.3	0.0	3.3	28.9	72.8	78.8
Z210	Inc. Z010(C), (Pol %S gp)	5272	16.55	15.95	24.3	19.0	82.7	0.0	3.9	21.5	62.1	72.6
03-FC1030-16	Inc. 01-FC1030-16 (A,aa)	4935	17.04	14.48	26.0	23.5	81.5	0.0	3.7	7.5	69.1	86.5
03-C37	susc. ck., Inc. U86-C37	4928	17.04	14.35	25.8	23.5	81.9	0.0	3.6	4.4	70.1	85.5
Lines with *Bvm* germplasm												
R336	RZM-ER-% R136, (C79-8)	5466	19.02	14.27	23.3	20.3	79.7	0.0	3.5	21.1	65.3	83.9
R340	RZM-ER-% R140	8580	26.96	15.83	26.3	23.5	82.0	0.0	3.4	27.1	64.3	82.0
R324/5	Inc. R824, (C79-2,-3; WB41,42)	4830	15.64	15.43	21.8	21.5	80.5	0.0	3.5	12.6	74.2	90.8
R324	Inc. R724, (C79-2, WB41)	5602	19.14	14.63	24.8	22.5	79.3	0.0	3.4	17.8	69.1	91.6
R325	Inc. R725, (C79-3, WB42)	5778	18.56	15.55	22.3	22.0	79.1	0.0	3.4	12.7	74.3	94.8
R337	Inc. R637, (C79-9, WB151)	6624	21.36	15.52	23.0	23.0	79.8	0.0	3.6	8.7	69.4	87.6
R641	RZM R541, R548, (C79-10, WB169)	5183	17.91	14.43	19.3	17.5	79.6	0.0	4.5	9.5	47.2	63.8
R642	RZM R542, R549, (C79-11, WB258)	6268	22.52	13.85	21.0	17.8	80.0	0.0	3.7	24.8	61.3	74.8

TEST 7104. HARTNELL FIELD EVALUATION OF GERMPLASM FOR REACTION TO IMPERIAL STRAIN OF BNYVV, SALINAS, CA, HARTNELL, 2004

(cont.)

Variety	Description	Sugar Yield			Stand Count	Harv Count	RJAP	Bolting	Rhizomania Resistance			
		Sugar Lbs	Beets Tons	Sucrose %	No.	No.	%	%	DI	%R(0-1)	%R(0-4)	%R(0-5)
Lines with *Bvm* germplasm (cont.)												
R740	RZM-ER R540%, R540-1, R551, (C79-#C)	7655	25.32	15.10	25.3	23.3	81.1	0.0	2.9	36.8	76.2	83.9
R840	RZM R740, (C79-#C)	6475	21.36	15.18	24.0	22.8	79.7	0.0	3.6	23.5	62.1	84.8
03-C37	susc. ck., Inc. U86-C37	4658	15.87	14.60	25.3	23.5	79.2	0.0	3.9	15.2	57.9	77.1
US H11	susc. ck., 10/14/02	3481	14.24	12.42	26.5	21.5	79.0	0.0	4.4	15.9	51.9	69.0
Beta 4430R	9/21/03	8611	29.29	14.82	26.0	24.8	83.9	0.0	2.3	53.6	83.7	91.0
Angelina	resist. ck., 2/25/04	10932	35.01	15.70	25.5	25.8	79.8	0.0	2.3	43.5	91.1	97.9
R318-6(Sp)	Inc. P118-6, (CP08)	5404	18.06	15.02	24.3	22.3	79.7	0.0	4.9	3.3	40.8	62.8
P207/8(Sp)	Inc. P007/8, (CP07)	7740	25.67	15.07	25.3	22.3	81.3	0.0	3.9	5.1	58.4	85.1
P329	PMR-RZM P229, (CP05)	7974	26.26	15.10	23.3	22.3	83.2	0.0	4.4	12.3	46.7	60.1
P330	PMR-RZM P230, (CP06)	7211	23.46	15.27	25.8	21.8	83.1	0.0	4.8	6.0	38.9	62.4
N312	PMR-RZM-NR N112, (CN12)	5288	17.62	14.98	22.3	21.8	82.8	0.0	4.5	7.0	48.4	71.0
N372	RZM-ER-NR N172, (CN72)	6047	20.66	14.27	21.5	19.8	80.1	0.0	5.4	2.4	30.6	42.6
R522 (Sp)	RZM R322(C), (C51)	5155	18.32	14.05	19.5	18.0	79.7	0.0	3.3	28.0	68.2	89.0
R021 (Sp)	RZM R926, RZM R927, (C26 x C27)	7474	25.09	14.90	25.0	23.5	81.6	0.0	3.6	13.2	63.8	82.9
R321	RZM R221, (C26 x C27)	5392	19.14	14.00	23.3	20.8	81.3	0.0	3.2	22.7	77.1	85.1
R926	RZM R826, (C26)	6754	23.34	14.48	21.3	20.0	80.9	0.0	3.4	19.6	71.9	86.4
R927	RZM R827, (C27)	7537	24.99	15.05	23.5	21.8	81.5	0.0	3.0	33.9	75.9	84.5
R720	RZM *B.vulgaris maritima*	2102	8.40	12.02	18.8	17.5	67.5	41.6	3.3	31.6	71.5	81.1
R423	Inc. R323 (RZM *B.maritima*)	3376	12.23	13.73	21.3	19.5	71.5	6.3	3.2	30.2	69.1	82.0
R423B	RZM PI518408-540610, *Bvm*	1341	6.54	10.27	21.3	18.8	59.5	47.8	2.8	35.9	79.2	83.3
R321	RZM R221, (C26 x C27)	6761	23.22	14.63	24.5	19.8	83.3	0.0	3.1	33.1	69.3	81.0
Y375	RZM Y275	4775	17.04	13.83	25.5	19.3	82.4	0.0	3.9	25.5	58.8	67.6
Y375-9	Inc. Y175-9	3958	14.86	13.45	26.3	20.8	81.0	0.0	4.0	13.4	60.1	77.1
Y175-13	Inc. Y175-13	4997	17.39	14.28	25.3	21.8	83.1	0.0	5.0	2.5	32.5	61.3

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TEST 7104. HARTNELL FIELD EVALUATION OF GERMPLASM FOR REACTION TO IMPERIAL STRAIN OF BNYVV, SALINAS, CA, HARTNELL, 2004

(cont.)

Variety	Description	Sugar Yield Sugar Lbs	Sugar Yield Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	RJAP %	Bolting %	Rhizomania Resistance DI	Rhizomania Resistance %R(0-1)	Rhizomania Resistance %R(0-4)	Rhizomania Resistance %R(0-5)
Lines with _Bvm_ germplasm (cont.)												
Y375-20	Inc. Y175-20	4142	14.82	13.95	23.5	21.5	77.6	0.0	5.3	2.9	25.0	52.6
Y367-5	RZM Y167-5 (SB x C51)	6771	22.52	14.85	21.0	20.5	82.8	0.0	3.3	41.4	69.8	73.5
Y367	RZM-ER-% Y167, (C67/2)	8029	25.91	15.45	24.3	23.0	82.6	0.0	3.0	30.6	80.5	89.4
Y277	RZM Y175-R136	6489	22.64	14.40	22.5	21.0	79.6	0.0	3.8	9.1	71.5	79.0
R343	RZM-ER-% R143	5694	18.91	15.07	22.0	19.8	83.2	0.0	3.3	17.3	72.1	85.3
R371	RZM-ER-% Y171	6526	21.36	15.18	23.3	21.8	81.7	0.0	3.9	13.9	60.3	75.3
R641	RZM R541, R548, (C79-10, WB169)	4732	17.86	13.35	16.0	16.3	78.9	0.0	3.9	13.4	52.5	85.7
R642	RZM R541, R549, (C79-11, WB258)	5713	21.24	13.55	20.8	19.8	79.6	0.0	4.0	11.7	56.9	71.7
R337	Inc. R637, (C79-9, WB151)	7884	25.32	15.57	25.8	24.5	79.4	0.0	2.8	26.5	84.8	98.0
R740	RZM-ER-% R540%, R540-1, R551	6724	22.76	14.73	23.3	22.5	80.7	0.0	3.3	22.5	70.2	89.0
R940	RZM-ER-% R740, (C79-#C)	8206	27.50	15.00	21.5	19.0	78.7	0.0	2.8	39.0	75.3	92.9
R926	RZM R826, (C26)	6059	21.24	14.30	23.0	21.8	78.4	0.0	2.9	42.5	75.8	82.1
R927	RZM R827, (C27)	6646	22.17	15.02	25.3	23.0	81.2	0.0	2.8	39.0	79.0	89.3
R021	RZM R926, R927, (C26xC27)	7480	26.02	14.45	22.0	21.3	82.6	0.0	3.3	26.7	68.4	84.2
Quantitative resistance												
R039	Inc. R539, (C39R)	10039	32.79	15.35	22.3	22.0	80.8	1.1	2.6	33.3	88.8	97.8
R647	RZM R547, (C47R)	9575	29.64	16.18	22.5	21.5	81.6	0.0	2.9	34.0	78.1	89.4
Multigerm, S^f,Aa populations												
2933	RZM %9933 (A,aa)	6539	22.17	14.67	23.8	23.0	82.1	0.0	4.8	11.2	38.5	56.4
3931	RZM 2931,1931aaxA, (C931)	6006	20.42	14.68	24.8	24.3	79.6	0.0	4.6	7.3	41.5	63.0
3941	RZM 2941,1941aaxA, (C941)	7757	25.79	15.02	25.0	24.0	84.5	0.0	3.9	9.2	59.6	80.7
2921	RZM 0921 (A,aa), (C51,C26,C27)	7534	24.97	15.05	24.3	23.5	81.8	0.0	3.2	14.6	75.5	90.6

TEST 7104. HARTNELL FIELD EVALUATION OF GERMPLASM FOR REACTION TO IMPERIAL STRAIN OF BNYVV, SALINAS, CA, HARTNELL, 2004

(cont.)

Variety	Description	Sugar Yield		Sucrose	Stand Count	Harv Count	RJAP	Bolting	Rhizomania Resistance			
		Sugar Lbs	Beets Tons	%	No.	No.	%	%	DI	%R(0-1)	%R(0-4)	%R(0-5)
Multigerm, S^f,Aa populations (cont.)												
0926	RZM-% 8926(Sp),(931xC51)	5835	20.09	14.52	24.3	21.5	81.7	0.0	3.9	13.6	62.5	79.1
Z325	RZM Z225, Z125(C)aa x A, (CZ225/2)	7322	23.92	15.23	22.5	20.3	82.7	0.0	4.5	9.4	44.4	67.1
CR311	RZM CR111,CR111(C)aa x A, (CR11)	7369	25.70	14.30	22.0	22.5	82.9	0.0	3.8	13.8	58.3	77.1
3927-4	RZM 2927-4 (A,aa),(C927-4)	6066	21.47	14.10	24.0	21.5	82.3	0.0	4.3	4.8	57.0	67.9
Monogerm populations and lines												
3812H5	C833-5HO x 6812 (C890-2,-3;WB41,42)	5009	18.05	13.75	22.0	20.0	80.0	1.3	4.8	9.7	42.0	60.9
3819H5	C833-5HO x 6819 (C890-9; WB151)	4560	16.68	13.70	21.8	18.3	82.3	0.0	4.6	7.4	46.4	62.6
2790H5	C833-5HO x 0790	5934	21.01	14.10	23.8	21.0	82.5	0.0	4.7	5.0	48.6	67.1
2848H5	C833-5HO x RZM,T-O 1848-#(C),(mm x C51)	5236	18.56	14.15	23.0	20.8	79.8	0.0	4.3	13.4	55.0	65.1
1848M	RZM 0848 (A,aa),(mm x C51)	5177	19.26	13.50	21.5	19.0	81.7	0.0	2.8	35.2	79.5	91.5
3869	RZM 1869(C)mmaa x A,(C869)	4878	18.32	12.90	22.5	20.5	81.7	0.0	4.4	11.3	52.5	65.7
3842	RZM 2842(C)mmaa x A,(C842)	3886	14.82	13.40	23.5	19.8	81.1	0.0	4.2	5.5	56.4	80.4
03-FC1015	RZM 02-FC1015mmaa x A	4319	16.57	12.68	22.0	19.3	77.3	0.0	4.7	5.5	45.6	66.4
Mean		6296.9	21.72	14.37	23.4	21.2	80.7	1.0	3.7	20.3	63.0	78.6
LSD (.05)		1656.7	5.35	1.40	4.0	4.6	4.8	4.3	1.1	20.0	23.4	21.7
C.V. (%)		18.9	17.70	7.00	12.2	15.7	4.2	303.7	22.3	71.0	26.7	19.8
F value		8.2**	7.54**	3.74**	1.7**	1.3*	3.5**	17.3**	3.4**	3.3**	3.2**	2.2**

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TEST 7104. HARTNELL FIELD EVALUATION OF GERMPLASM FOR REACTION TO IMPERIAL STRAIN OF BNYVV,
SALINAS, CA, HARTNELL, 2004
(cont.)

| Variety | Description | Sugar Yield | | Sucrose | Stand | Harv | RJAP | Bolting | Rhizomania Resistance | | | |
		Sugar Lbs	Beets Tons	%	Count No.	Count No.	%	%	DI	%R(0-1)	%R(0-4)	%R(0-5)

NOTES: Test 7104 was grown on Hartnell College field. In 2003, soil from Imperial Valley (Rockwood 156) field was run thru planter openers into beds and grown from June to October, 2003. Rhizomania was evident but mild. In 2004, this isolated field area was prepared for sugarbeet trials. Test 7104 was planted into beds in early May. The trial area was frequently sprinkler irrigated to promote rhizomania development. Throughout the season, the growth and appearance of most entries were typical of rhizomania infection. In several rz1rz1 entries, systemic infection was evident. The field was treated for black aphids and powdery mildew controlled, but otherwise, there were few pest or disease problems noted. On November 18, 2004, test 7104 was hand-harvested. Roots were lifted, laid out, and scored for rhizomania, topped, bagged, washed, weighed, and run through the sugar lab.

Rhizomania was scored on a scale of 0 to 9, where 0 = very smooth roots with no evidence of disease to 9 = dead due to rzm or root rot from rzm. The level of infection appeared moderate. Some roots showed typical rzm symptoms, some showed bearding on lateral roots only, and some appeared to have escaped early infection. Symptoms were a little different than the usual rzm expression at Salinas. The susceptible checks had more or less typical bearding. The Rz1 entries generally showed less severe bearding, but most showed moderate to severe lower tap root discoloration and vascular necrosis. Tip rot was fairly common in Rz1 roots even without significant bearding. Under the moderate climate conditions of Salinas, only a few of these roots died and the relative differences between stand counts and harvested roots may reflect early death. It is suspected that similar roots in the Imperial Valley under May-July conditions would show greater rotting and death. It appeared that escapes and partial escapes (late infection) were still a concern in this field trial. Thus, in 2005, tests in this location should be much more severe.

Rhizomania reaction is shown as DI (disease index), average individual root rating, and %R (% resistant). %R was calculated for roots only scored as 0-1 (%R,0-1), scored as 0-4 (%R, 0-4), and scored as 0-5 (%R, 0-5). Because of late infection, it was felt that the %R (0-1) scores might more accurately show frequency of resistant plants and disease reaction. DI also appears to be a good indicator of disease reaction among entries.

Results: This screening trial with 96 entries and only 4 replications and only moderate disease frequency and level is not very definitive. It appeared that Rz1 still may have given partial control of the rhizomania in this field. Rz2 entries may have also been better, but Rz2 was in less advanced, less vigorous backgrounds. Some germplasm lines with B.vulgaris subsp. maritima (Bvm) sources also appeared to be somewhat more resistant.

TEST 7104. HARTNELL FIELD EVALUATION OF GERMPLASM FOR REACTION TO IMPERIAL STRAIN OF BNYVV,
SALINAS, CA, HARTNELL, 2004

(cont.)

| | | Sugar Yield | | Stand | Harv | | | | Rhizomania Resistance | |
| Variety | Description | Sugar Lbs | Beets Tons | Sucrose % | Count No. | Count No. | RJAP % | Bolting % | DI %R(0-1) %R(0-4) | %R(0-5) |

To some extent, lines with repeated selection for resistance to rhizomania appeared to be more tolerant, with
or without the *Rz1* and *Rz2* major genes. This was most evident in C39R (R039) and C47R (R647) that have only
quantitative (additive) resistance to rhizomania. C39R and C47R were developed after 4 t0 6 cycles of
phenotypic recurrent selection for resistance to rhizomania and are not known to have major gene resistance.

Coefficients of correlation (r) were calculated:

	%Resist (0-1)	%Resist (0-4)	%Resist (0-5)	HC	SY	RY	%S
Disease Index	-0.81**	-0.96**	-0.89**	-0.20**	-0.45**	-0.46**	-0.15**
% Resistant (0-1)				0.06NS	0.37***	0.39***	0.04NS
% Resistant (0-4)				0.23**	0.43***	0.43***	0.16**
% Resistant (0-5)				0.24**	0.40***	0.40***	0.18**
Harvest Count					0.37**	0.35**	0.26**
Sugar Yield						0.96**	0.57**
Root Yield							0.33**

TEST 2304. EVAL. OF LINES WITH RESISTANCE TO POWDERY MILDEW, SBCN, AND/OR RHIZOMANIA, SALINAS, CA, 2004

32 entries x 6 reps., sequential
1-row plots, 11 ft. long

Planted: March 22, 2004
Harvested: October 13, 2004

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	RJAP %	Bolting %	Powdery Mildew Scores 8/13	9/07	10/8	Mean
Checks											
03-C37	Inc. U86-37, 99-C37	13513	41.39	16.30	141	81.8	0.0	8.8	6.8	8.0	7.5
P327	PMR-RZM P227, (CP03)	14143	44.62	15.88	142	81.0	0.0	2.8	2.5	7.3	3.3
P328	PMR-RZM P228, (CP04)	16536	50.66	16.35	145	81.5	0.0	2.2	1.8	3.5	1.9
US H11	11/3/99	14545	46.36	15.63	153	83.5	0.0	9.0	8.0	8.0	8.1
R378	RZM-ER-% R178	16652	49.05	16.97	142	80.5	0.0	6.3	4.3	5.3	4.7
P329	PMR-RZM P229, (CP05)	16646	50.39	16.57	147	82.3	0.0	1.7	1.0	1.3	1.1
P330	PMR-RZM P330, (CP06)	16692	49.33	16.90	133	82.4	0.0	2.8	1.3	1.8	1.7
Angelina	2002	19685	57.38	17.15	145	82.9	0.0	9.0	8.2	7.8	8.1
P227	PMR-RZM-NB P027-#(C),(CP03)	13313	42.73	15.55	144	80.3	6.3	5.2	3.8	6.8	4.6
P601	PMR P401 (WB97,242)	13880	43.00	16.17	133	81.1	1.2	3.7	2.7	3.8	3.1
P229	PMR-RZM-NB P029-#(C),(CP05)	16902	49.71	17.02	133	82.4	0.0	2.5	2.3	2.2	2.2
P230	PMR-RZM-NB P030-#(C),(CP06)	17267	51.33	16.82	144	80.1	0.0	4.2	2.7	3.3	3.1
P318-6(Iso)	PMR-RZM P118-6,(CP08)	17022	52.14	16.33	150	80.8	0.0	4.0	2.0	3.3	2.6
P318-6(Sp)	Inc. P118-6,(CP08)	15447	47.84	16.15	145	80.0	0.0	5.2	3.2	4.3	3.9
P207/8(Iso)	RZM-PMR-NR P007/8,(CP07)	17665	51.74	17.05	148	82.2	0.0	1.7	0.8	1.8	1.3
P207/8(Sp)	Inc. P007/8,(CP07)	15820	48.11	16.45	152	81.5	0.0	1.7	1.0	2.0	1.3
US H11	11/3/99	14095	45.42	15.55	155	84.1	0.0	9.0	8.0	7.8	8.1
P318-6H5	C833-5CMS x P118-6	18250	53.66	17.02	145	81.4	0.0	6.8	5.2	5.2	5.2
P207/8H5	C833-5CMS x P007/8	16936	49.05	17.27	148	80.7	0.0	3.8	3.3	3.3	3.2
Y391	RZM-ER-% Y191	16359	46.23	17.67	148	83.0	0.0	4.7	3.2	3.2	3.3
N112	NR-RZM P912 (A,aa)	15709	47.30	16.62	142	80.9	0.0	4.7	2.8	4.3	3.5
N312	PMR-RZM-NR N112	16613	49.86	16.68	153	81.1	0.0	2.0	1.2	1.7	1.4
N372	RZM-ER-NR N172	15772	47.44	16.67	142	80.6	0.0	3.0	4.2	3.5	3.2
Y375	RZM Y275	15918	48.38	16.48	142	80.9	0.0	7.0	5.7	4.5	5.1

TEST 2304.   EVAL. OF LINES WITH RESISTANCE TO POWDERY MILDEW, SBCN, AND/OR RHIZOMANIA, SALINAS, CA, 2004

(cont.)

| Variety | Description | Acre Yield | | Sucrose | Beets/100' | RJAP | Bolting | Powdery Mildew Scores | | | |
		Sugar Lbs	Beets Tons	%	No.	%	%	8/13	9/07	10/8	Mean
Checks (cont.)											
R321	RZM R221	15804	48.11	16.38	145	84.0	0.0	6.7	5.5	5.8	5.3
R336	RZM-ER-% R136	13311	42.47	15.70	144	81.1	0.0	8.5	6.8	5.2	6.3
R039	Inc. R539, (C39R)	13449	43.57	15.40	133	83.2	0.0	3.5	3.7	3.8	2.9
Z210	Inc. Polish %s (C)	14625	39.24	18.80	148	81.6	0.0	8.0	7.8	6.5	7.2
N325	RZM N224(C) (g)	14500	44.35	16.35	144	81.6	0.0	5.3	5.7	4.2	4.8
N369	RZM N269-#(C) (g)	12615	38.70	16.30	156	81.7	0.0	7.3	7.7	5.0	6.5
02-WB97	Inc. WB97, (C37 x WB97)	10282	34.94	14.68	150	80.7	45.5	1.7	1.7	3.0	1.9
02-WB242	Inc. WB242, (C37 x WB242)	10422	35.05	14.88	138	78.5	32.6	2.5	2.0	4.3	2.5
Mean		15324.6	46.55	16.43	144.9	81.6	2.7	4.9	4.0	4.4	4.0
LSD (.05)		1830.6	5.30	0.78	13.3	2.7	4.3	1.6	1.3	1.3	1.1
C.V. (%)		10.5	9.98	4.15	8.0	2.9	139.5	28.8	29.0	25.9	24.9
F value		10.1**	7.22**	8.34**	1.6*	1.6*	40.9**	19.4**	25.7**	18.0**	28.3**

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TEST 2204. EVALUATION OF FULL-SIB PROGENIES WITH WB97 & 242 GERMPLASM, SALINAS, CA, 2004

48 entries x 3 reps., sequential
1-row plots, 11 ft. long

Planted: March 22, 2004
Harvested: October 13, 2004

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	RJAP %	Powdery Mildew Scores 8/13	9/07	10/8	Mean
Checks										
US H11	10/4/02	14528	44.62	16.27	148	82.7	9.0	7.7	7.7	8.2
P327	PMR-RZM P227, (CP03)	15902	49.72	16.00	142	80.9	2.7	2.0	7.7	3.4
P328	PMR-RZM P228, (CP04)	18346	57.44	16.00	158	81.6	2.3	1.0	3.0	1.9
P329	PMR-RZM P229, (CP05)	17437	52.95	16.47	139	81.8	1.0	0.7	1.0	0.9
P330	PMR-RZM P330, (CP06)	18797	56.12	16.73	139	80.0	2.3	1.3	1.3	1.7
03-C37	Inc. U86-37, 99-C37	12748	41.93	15.10	145	84.6	9.0	7.7	9.0	7.7
P318-6	PMR-RZM P118-6, (CP08)	16129	49.99	16.13	142	80.1	2.7	1.3	2.7	2.1
P307/8	PMR-RZM P207/8, (CP07)	17583	50.26	17.50	121	82.8	1.7	0.3	1.3	1.3
P207/8 (Iso)	RZM-PMR-NR P007/8, (CP07)	18319	54.02	16.97	139	82.9	0.3	0.3	1.3	0.7
Roberta	3/25/03	18942	59.40	15.93	161	84.9	8.0	5.3	6.3	6.3
FS's from P207/8 PX										
P307-301	PMR-RZM P207/8 PX	18613	54.56	17.10	139	80.3	2.3	1.3	3.0	1.8
-302		18860	55.10	17.10	152	84.2	0.0	0.3	3.7	0.8
-303		18975	54.83	17.30	152	81.6	0.0	0.0	1.7	0.3
-304		18096	55.90	16.17	136	79.7	0.0	0.3	6.0	1.2
-305		18070	52.68	17.13	145	82.5	0.0	0.0	0.7	0.1
-306		16993	51.07	16.63	139	80.4	0.7	0.7	1.3	0.9
-307		16789	52.41	16.10	148	82.7	0.7	0.7	1.3	1.0
-308		15536	45.69	17.00	139	82.1	0.3	0.3	1.0	0.4
-309		15691	48.38	16.20	133	79.0	1.0	0.7	1.3	0.8
-310		17003	51.15	16.67	121	82.1	0.0	0.0	0.3	0.1
-311		17679	56.17	15.73	121	83.1	0.0	0.0	0.7	0.1
-312		15882	53.22	14.90	127	81.3	0.0	0.0	2.0	0.5
-313		16513	51.60	15.97	139	82.0	0.0	0.0	0.7	0.1
-314		18550	57.78	16.03	142	81.7	0.0	0.0	1.7	0.3
-315		17741	54.02	16.43	139	80.8	1.0	0.7	1.0	0.9

TEST 2204. EVALUATION OF FULL-SIB PROGENIES WITH WB97 & 242 GERMPLASM, SALINAS, CA, 2004

(cont.)

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	RJAP %	Powdery Mildew Scores 8/13	Powdery Mildew Scores 9/07	Powdery Mildew Scores 10/8	Powdery Mildew Scores Mean
FS's from P229PX										
P329-301	PMR-RZM P229PX	20407	61.01	16.83	139	80.6	1.0	1.3	1.7	1.7
-302		16294	52.57	15.50	118	84.3	0.0	0.0	1.0	0.2
-303		17352	53.75	16.13	139	82.2	0.0	0.0	0.7	0.1
-304		18293	57.78	15.87	139	82.6	0.0	0.0	0.7	0.1
-305		17056	51.07	16.70	145	81.2	0.0	0.0	0.0	0.0
-306		16863	49.72	16.97	133	81.9	1.3	1.3	1.0	1.4
-307		18350	56.01	16.43	130	82.7	1.0	0.3	1.0	0.7
-308		18760	56.58	16.60	127	83.3	0.7	0.3	1.0	0.7
-309		18694	56.44	16.50	142	81.9	0.0	0.0	0.3	0.1
-310		18525	55.90	16.60	124	83.1	1.3	0.7	1.3	1.3
-311		18269	54.83	16.67	127	83.2	0.0	0.0	0.7	0.1
FS's from P230PX										
P330-301	PMR-RZM P230PX	21118	61.01	17.30	139	81.8	2.7	1.3	1.7	1.8
-302		18674	54.29	17.23	139	78.3	2.7	1.0	2.0	1.8
-303		18929	55.63	17.00	133	83.5	2.0	0.3	2.0	1.3
-304		18280	52.41	17.47	148	80.3	6.7	2.3	4.7	4.8
-305		17370	52.68	16.50	127	82.1	3.3	0.3	1.3	1.3
-306		16586	49.16	16.90	139	81.6	4.3	3.0	3.3	3.3
-307		18864	56.55	16.70	142	81.2	4.7	4.3	4.3	3.8
-308		18026	51.07	17.67	152	80.3	8.3	6.3	6.3	6.7
-309		17779	54.25	16.37	127	80.5	5.7	1.3	3.0	2.8
-310		18306	54.29	16.87	124	82.3	2.3	0.3	1.7	1.2
-311		19566	58.33	16.77	103	79.9	1.7	0.3	1.0	0.8
-312		17196	52.22	16.47	133	80.3	7.0	4.7	3.0	5.2
Mean		17693.3	53.51	16.53	137.2	81.8	2.1	1.3	2.3	1.8
LSD (.05)		2722.7	7.83	1.07	19.5	3.1	1.6	1.2	1.6	1.0
C.V. (%)		9.5	9.03	4.00	8.8	2.3	46.9	57.5	41.1	35.0
F value		2.3**	1.92**	2.39**	2.5**	1.7*	21.1**	20.9**	15.1**	33.3**

TEST 4304. EVALUATION OF ROOT KNOT NEMATODE (RKN) GERMPLASM LINES, SALINAS, 2004, BLOCK 2

12 entries x 4 reps., sequential
1-row plots, 11 ft. long

Planted: May 3, 2004
Harvested: December 2, 2004

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Bolting %	Beets/100' No.	RJAP %
Checks							
03-C37	susc. ck., Inc. U86-37	10681	30.11	17.73	0.0	182	80.4
R378	RZM-ER-% R178	15404	40.72	18.90	0.0	170	82.0
US H11	susc. ck., 10/14/02	11378	35.90	15.88	0.0	202	83.0
WB66 source							
M66	M66 (1998), 640 g	4592	18.45	12.18	53.5	168	75.0
M6-1	M6-1 (2000), 983 g	8063	21.86	18.10	0.0	164	83.8
M6-2	M6-2 (2002), 980 g	12095	34.10	17.75	0.0	207	82.7
M6-3	M6-2 (2003), 459 g	11211	31.69	17.70	0.0	202	81.5
WB258 source							
Mi-1	Mi-1 (1998), 100g	9122	25.72	17.58	47.5	168	82.7
M1-2	M1-2 (2000), 811 g	9637	26.68	18.07	56.5	164	83.5
M1-3	M1-3 (2002), 963 g	13501	37.37	18.08	0.0	184	82.8
M1-3	M1-3 (2003), 751 g	15476	42.92	17.98	0.0	216	83.2
M1-4	M1-4 (2003), 284 g	14458	40.36	17.92	0.0	180	82.5
Mean		11301.5	32.16	17.32	13.1	183.9	81.9
LSD (.05)		2785.5	7.06	1.43	9.4	23.5	5.2
C.V. (%)		17.1	15.27	5.74	49.6	8.9	4.4
F value		11.0**	10.12**	12.57**	53.7**	5.1**	1.7NS

TESTS 1104-4404. EVALUATION OF MULTIGERM PROGENY LINES WITH AND WITHOUT RHIZOMANIA, SALINAS, CA, 2004

Planted: March 22 & May 3, 2004
Harvested: September 23 & December 2, 2004

(48 entries x 4 reps.) x 2 tests, sequential
1-row plots, 11 ft. long

Variety	Description	Acre Yield		Sucrose	Beets/100'	RJAP	Powdery Mildew Scores			
		Sugar Lbs	Beets Tons	%	No.	%	8/23	8/30	9/07	Mean
Checks										
Y391	RZM-ER-% Y191	15022	42.03	17.98	180	83.8	2.3	2.0	1.8	2.0
Roberta	3/25/03	15744	47.77	16.54	185	84.6	4.3	3.5	3.8	3.8
MM,S^f Aa,S$_1$ progeny lines										
2930-19	RZM 1930-19aa x A, (C930-19)	14061	40.53	17.41	151	83.9	4.0	2.3	2.5	2.9
Z325-9	RZM Z225-9 (A,aa),(CZ25-9)	13459	36.08	18.77	158	81.9	2.0	1.8	2.0	1.9
2930-35	RZM 1930-35aa x A, (C930-35)	14106	37.93	18.67	173	83.1	8.0	6.3	6.5	6.9
3927-4	RZM 2927-4 (A,aa), (C927-4)	13376	41.12	16.49	176	82.2	7.3	4.5	4.5	5.4
3931-56	RZM 1931-56aa x A	13397	37.79	17.79	159	83.0	2.3	1.3	1.3	1.6
Z331-14	RZM Z131-14aa x A	14929	39.61	18.94	175	82.8	5.5	3.3	4.5	4.4
Z325-105	Inc. Z125-105 (A,aa)	12032	33.86	17.88	183	82.7	5.8	5.8	6.8	6.1
Z325-109	Inc. Z125-109 (A,aa)	11097	31.65	17.65	179	83.2	5.0	3.3	4.5	4.3
03-FC1030-15	03-FC1030-15 (A,aa)	13519	36.48	18.59	184	82.8	4.5	2.3	3.3	3.3
03-FC1030-16	03-FC1030-16 (A,aa)	14362	40.51	17.81	184	83.3	4.3	3.0	3.5	3.6
CR310-14-2	RZM CR110-14-2 (A,aa)	9523	28.66	16.66	177	80.9	9.0	6.8	7.0	7.6
CR311-6	Inc. CR111- 6 (A,aa)	14385	41.88	17.23	175	82.2	4.3	3.3	3.3	3.6
CR311-41	Inc. CR111-41 (A,aa)	12996	37.79	17.31	168	83.4	8.5	7.3	7.3	7.7
CR311-88	Inc. CR111-88 (A,aa)	16010	47.06	17.05	181	82.7	6.5	4.5	5.0	5.3
2933-14	Inc. 0933-14 (A,aa)	12388	34.57	18.04	181	83.2	1.0	0.0	0.8	0.6
3933-107	Inc. 1933-107 (A,aa)	13389	39.40	17.14	180	83.3	1.8	1.3	2.0	1.7
3933-113	Inc. 1933-113 (A,aa)	13858	39.71	17.50	184	83.6	2.3	1.0	1.5	1.6
3933-118	Inc. 1933-118 (A,aa)	14661	40.99	18.00	164	84.1	7.0	3.5	4.3	4.9
2936-16	0936-16aa x A	13215	36.22	18.25	180	80.5	3.3	2.5	3.0	2.9
3931-120	Inc. 1931-120 (A,aa)	12549	37.09	17.00	171	80.6	6.8	3.8	4.3	4.9
2941-20	Inc. 0941-20 (A,aa)	13373	38.46	17.49	164	82.0	6.3	3.3	3.0	4.2
3941-107	Inc. 1941-107 (A,aa)	12946	35.58	18.31	181	83.5	8.5	7.3	7.5	7.8
3941-112	Inc. 1941-112 (A,aa)	14501	39.81	18.27	181	82.9	3.8	2.8	3.5	3.3

TESTS 1104-4404. EVALUATION OF MULTIGERM PROGENY LINES WITH AND WITHOUT RHIZOMANIA, SALINAS, CA, 2004 (cont.)

Variety	Description	Acre Yield			Beets/ 100'	RJAP	Powdery Mildew Scores			
		Sugar Lbs	Beets Tons	Sucrose %	No.	%	8/23	8/30	9/07	Mean
MM,S^{ss}S^s,FS progeny lines										
P318-6 Sp	Inc. P118-6	13862	40.81	17.02	184	81.5	6.5	5.0	6.0	5.8
R280/2-9	Inc. R080/2-9	13918	38.78	18.13	167	82.7	8.0	6.8	7.3	7.3
R280-6	Inc. R080-6	15441	44.75	17.40	186	82.9	5.5	3.5	4.8	4.6
R380-21	RZM R180-21	14217	40.61	17.52	175	83.0	7.0	5.3	6.0	6.1
Y390-40	Inc. Y190-40	14639	40.51	18.09	190	80.6	8.0	6.5	7.8	7.4
Y390-43	Inc. Y190-43	13174	37.39	17.75	181	82.5	4.0	1.8	2.3	2.7
Y390-83	Inc. Y190-83	12872	36.18	17.84	181	82.9	5.5	4.3	4.5	4.8
Y390-98	Inc. Y190-98	14517	42.41	17.24	180	83.5	2.0	1.5	1.8	1.8
R378-6	RZM R178-6	13594	38.96	17.54	153	83.4	4.3	3.5	4.3	4.0
Y368-8	RZM Y168-8	13875	38.85	18.00	180	84.1	3.5	2.8	4.0	3.4
Y269-8	Inc. Y069-8	13399	35.88	18.69	183	82.2	5.8	4.0	4.8	4.8
Y269-39	Inc. Y069-39	15055	41.02	18.39	175	82.6	3.3	1.5	2.0	2.3
R381-22	RZM R181-22	14294	39.30	18.24	182	83.9	6.0	3.8	4.0	4.6
R243-14	Inc. R043-14	15442	44.24	17.52	179	84.8	3.0	1.5	2.0	2.2
R376-89-10	Inc. R376-89-10	11115	29.87	18.67	178	82.8	2.3	1.5	2.5	2.1
Y367-5	RZM Y167-5	15054	42.36	17.88	177	83.5	2.5	1.3	1.8	1.8
Y267-21	Inc. Y067-21	13344	36.78	18.14	185	83.7	1.8	1.3	2.5	1.8
Y267-34	Inc. Y067-34	14032	39.51	17.94	174	82.9	1.5	0.8	2.0	1.4
Y271-14	Inc. Y071-14	13414	38.40	17.50	173	81.7	5.5	4.3	4.8	4.8
Y375-9	Inc. Y175-9	13829	40.82	17.13	181	83.7	4.8	2.8	3.3	3.6
Y375-13	Inc. Y175-13	14000	39.81	17.71	173	83.1	3.8	2.0	3.0	2.9
Y375-20	Inc. Y175-20	13936	40.26	17.44	172	84.2	4.0	1.8	3.3	3.0
P318-6	PMR-RZM P118-6	13454	40.61	16.76	175	82.1	2.3	1.0	1.5	1.6
Mean		13737.0	38.97	17.74	176.2	82.9	4.7	3.2	3.8	3.9
LSD (.05)		2065.8	5.74	0.77	19.6	2.8	1.9	1.5	1.8	1.4
C.V. (%)		10.8	10.64	3.11	8.2	2.4	29.6	32.8	34.3	26.1
F value		5.2**	6.30**	9.51**	2.8**	2.0**	9.8**	12.6**	8.1**	14.4**

TEST 1904-5204. EVALUATION OF MONOGERM LINES AND POPULATIONS WITH AND WITHOUT RZM, SALINAS, CA, 2004

Planted: March 22 & May 3, 2004
Harvested: October 12 & November 29, 2004

(48 entries x 4 reps.) x 2 tests, sequential
1-row plots, 11 ft. long

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	RJAP %	Powdery Mildew Scores 8/23	9/07	Mean
Checks									
2833-5NB	NB-RZM-% 0833-5-(Sp)aa x A	11803	33.36	17.71	178	79.3	7.5	6.5	7.0
99-C790-68	Inc. U88-790-68	10445	31.14	16.81	168	81.2	4.0	3.3	3.6
02-C790-15	Inc. 00-C79-15	13842	42.63	16.33	180	82.1	3.0	3.0	3.0
02-C790-15CMS	99-C790-68CMS x 00-C790-15	13919	41.93	16.65	164	82.1	5.3	4.8	5.0
0546	Inc. 97-C546, (C546)	9326	29.77	15.63	170	80.7	5.3	2.5	3.9
0562	Inc. 97-C562, (C562)	8550	27.72	15.51	173	82.8	6.3	4.0	5.1
2833-5(Sp)	RZM,T-O 1833-5-#(C)mmaa x A	12817	36.20	17.69	157	81.1	7.0	6.5	6.8
2833-5HO(Sp)	1833-5HOxRZM,T-O 1833-5-#(C)mmaa x A	12186	34.77	17.48	166	79.5	7.3	6.3	6.8
FC monogerm populations									
03-FC124	RZM 02-FC124mmaa x A	14699	41.02	17.99	155	82.4	6.8	4.3	5.5
03-FC124HO	RZM 02-FC124HO x A	13876	39.97	17.42	158	79.8	6.3	4.0	5.1
03-FC1015HO	RZM 02-FC1015HO x A	13283	37.69	17.65	168	81.8	7.5	6.0	6.8
03-FC1015	RZM 02-FC1015mmaa x A	12900	37.09	17.42	165	80.8	7.3	6.5	6.9
Monogerm lines, CMS's and F$_1$CMS's									
3837-6	RZM,T-O 2837-6-#(C)(A,aa)	12010	35.93	16.81	176	78.7	7.3	5.5	6.4
3837-6HO	0837-6H5 x RZM,T-O 2837-6-#(C)(A,aa)	15107	44.04	17.24	170	80.3	7.0	5.3	6.1
03-FC1014-22H5	C833-5CMS x 01-FC1014-22	15837	43.54	18.26	161	79.9	6.5	4.8	5.6
03-FC1014-22	Inc. 01-FC1014-22 (A,aa)	12462	34.46	18.13	189	80.0	7.3	5.5	6.4
03-FC123-31	Inc. 01-FC123-31 (A,aa)	11979	34.36	17.52	181	80.6	7.5	6.3	6.9
03-FC123-31H5	C833-5CMS x 01-FC123-31	14986	42.03	17.90	169	80.3	8.3	8.0	8.1
3869-24H5	C833-5CMS x 1869-24 (A,aa)	14946	44.04	17.00	167	79.6	7.0	5.5	6.3
3869-24	Inc. 1869-24 (A,aa)	12592	38.26	16.52	185	81.7	7.0	5.8	6.4
3869-27	Inc. 1869-27 (A,aa)	13920	43.54	16.10	184	82.2	6.8	5.0	5.9
3869-27H5	C833-5CMS x 1869-27	15273	44.44	17.23	165	80.9	7.0	6.5	6.8
3869-30H5	C833-5CMS x 1869-30	14500	41.32	17.61	174	81.2	7.0	4.3	5.6
3869-30	Inc. 1869-30 (A,aa)	13098	38.50	17.08	182	81.2	6.8	6.0	6.4

(cont.)

Variety	Description	Acre Yield		Sucrose	Beets/100'	RJAP	Powdery Mildew Scores		
		Sugar Lbs	Beets Tons	%	No.	%	8/23	9/07	Mean
Nematode resistant monogerms									
N365-31HO	RZM N265-31HO(g) x RZM N265-31(g)	10595	35.49	15.13	150	83.2	6.5	4.5	5.5
N365-9MHO	RZM N265-9HO(g) x RZM N265-9(g)	12991	44.31	14.79	148	81.3	8.3	7.0	7.6
N369	RZM N269-#(C)(g)	11532	35.07	16.50	174	79.1	8.0	6.3	7.1
N369HO	RZM N265-31HO(g) x RZM N269-#(C)(g)	13880	44.36	15.73	149	81.7	7.8	5.8	6.8
N365HO	RZM N265HO(g) x RZM N265(g)	10661	37.22	14.41	125	81.5	7.3	5.0	6.1
N365	RZM N265(g)	10798	37.69	14.36	168	81.8	7.8	5.5	6.6
N366	RZM N266(C)(g)	11645	39.61	14.76	177	77.9	6.5	4.5	5.5
N367	RZM N267(g)	12482	40.52	15.52	178	82.0	4.3	2.3	3.3
Monogerm populations									
3842	RZM 2842(C)mmaa x A	12318	36.28	17.04	170	80.1	7.0	5.8	6.4
3842HO	2842HO x A	12973	37.49	17.38	176	80.9	7.0	5.5	6.3
3842H5	C832-5NBHO x A	12747	36.51	17.49	175	79.5	6.8	5.3	6.0
3842H50	C790-15CMS x A	14566	41.89	17.42	169	82.3	6.0	5.8	5.9
3869	1869(C)mmaa x A	14556	43.40	16.79	176	82.6	6.5	6.0	6.3
3869HO	1869HO x A	15496	45.66	17.04	181	81.6	6.0	5.5	5.8
3869H5	C833-5NBHO x A	15140	43.41	17.45	170	81.0	5.8	4.8	5.3
3869H50	C790-15CMS x A	15849	46.87	17.02	185	81.9	5.3	3.8	4.5
2790	0790mmaa x A	13561	41.93	16.16	175	81.7	5.0	2.5	3.8
2848	RZM,T-O 1848-#(C)mmaa x A	13730	39.16	17.64	162	82.4	5.8	4.0	4.9
3849m	RZM 2251-2255(C)mmaa x A	15092	42.84	17.69	162	80.8	7.3	5.3	6.3
3849M	RZM 2251-2255(C)Maa x A	15762	43.04	18.34	176	80.5	5.8	5.5	5.6
3812	Inc. 6812M (A,aa)	11724	37.90	15.60	181	81.9	7.0	6.3	6.6
3812H5	C833-5CMS x A	15509	44.68	17.40	167	81.0	7.3	6.5	6.9
3819H5	C833-5CMS x A	15486	46.44	16.75	164	78.4	4.8	3.5	4.1
3819m	Inc. 6819 (A,aa)mm	15757	46.26	17.04	170	81.6	4.3	3.5	3.9

TEST 1904-5204. EVALUATION OF MONOGERM LINES AND POPULATIONS WITH AND WITHOUT RZM, SALINAS, CA, 2004

(cont.)

Variety	Description	Acre Yield			Beets/		Powdery Mildew		
		Sugar	Beets	Sucrose	100'	RJAP	Scores		
		Lbs	Tons	%	No.	%	8/23	9/07	Mean
Mean		13316.8	39.70	16.82	169.5	81.0	6.5	5.1	5.8
LSD (.05)		2121.2	5.92	1.15	21.9	3.7	2.0	2.0	1.7
C.V. (%)		11.4	10.66	4.89	9.7	3.2	21.8	27.9	21.3
F value		11.3**	9.22**	12.33**	4.0**	1.7*	2.6**	3.1**	3.5**

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TEST 604. PERFORMANCE HYBRIDS NOT INOCULATED WITH BYV, SALINAS, CA, 2004

24 entries x 8 reps., RCB(e)
1-row plots, 22 ft. long

Planted: March 19, 2004
Harvested: October 14, 2004
Not BYV inoculated

| Variety | Description | Acre Yield | | Sucrose | Beets/ 100' | RJAP | Powdery Mildew |
		Sugar Lbs	Beets Tons	%	No.	%	9/27
Checks							
Beta 4001R	8/25/03	17591	51.24	17.16	159	85.9	2.6
Phoenix	9/12/03	15922	49.38	16.13	158	87.6	4.9
HH142	9/12/03	15218	43.48	17.50	155	85.4	4.3
Beta 4430R	8/21/03	17454	51.16	17.06	158	87.2	3.9
Experimental hybrids							
Y291H5	C833-5CMS x RZM Y191	14577	41.52	17.56	151	84.2	4.0
Y391H50	C790-15CMS x RZM-ER-% Y191	15466	44.40	17.42	159	86.3	3.0
R376-89-4H50	C790-15CMS x R176-89-4	14767	42.98	17.16	165	86.2	4.4
R381-22H5	C833-5CMS x RZM R181-22	14673	41.27	17.79	151	85.2	2.8
Y375H50	C790-15CMS x RZM Y275	15499	45.10	17.17	161	84.6	4.3
Y392H50	C790-15CMS x RZM Y292	15079	44.04	17.11	165	85.1	4.3
Z210H5	C833-5CMS x Z010(C)	14824	39.30	18.85	148	84.9	5.3
2930-35H5	C833-5CMS x RZM 1930-35	14985	41.79	17.94	154	83.4	4.0
2930-19H5	C833-5CMS x RZM 1930-19	14582	41.62	17.50	148	84.6	3.5
R380-21H5	x RZM R180-21	15937	45.10	17.69	153	85.2	5.0
Y368-8H5	x RZM Y168-8	14841	42.66	17.39	153	84.3	3.3
R378-6H5	x RZM R176-6	15245	44.14	17.29	153	83.9	4.5
R378H5	C833-5CMS x RZM R178	14986	42.70	17.56	156	83.7	3.8
Y367-5H5	x RZM Y167-5	14398	40.41	17.84	157	85.8	3.0
P318-6H5	x P118-6	15683	45.05	17.38	156	83.9	2.9
3931-56H5	x RZM 1931-56	16249	45.85	17.71	153	84.6	2.8
3931H5	x RZM 2931,1931	15535	44.54	17.45	155	83.2	3.9
3941H5	x RZM 2941,1941	15246	43.69	17.44	154	84.5	3.8
3942H5	x RZM 2942	13766	38.68	17.80	153	84.0	3.3
Z325H5	x RZM Z225,Z125(C)	14958	41.62	17.94	147	83.7	4.6

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TEST 604. PERFORMANCE HYBRIDS NOT INOCULATED WITH BYV, SALINAS, CA, 2004

(cont.)

Variety	Description	Acre Yield		Sucrose	Beets/ 100'	RJAP	Powdery Mildew
		Sugar Lbs	Beets Tons	%	No.	%	9/27
Mean		15311.7	43.82	17.49	155.1	536.2	3.8
LSD (.05)		1095.2	3.07	0.48	8.9	1.8	1.0
C.V. (%)		7.3	7.12	2.79	5.8	2.1	25.6
F value		4.9**	8.49**	7.63**	2.2**	3.4**	4.9**

TEST 704. CYST NEMATODE COMMERCIAL AND EXPERIMENTAL HYBRIDS UNDER NONDISEASED CONDITIONS, SALINAS, 2004

24 entries x 8 reps., RCB(e)
1-row plots, 22 ft. long

Planted: March 19, 2004
Harvested: September 29, 2004

Variety	Resistance Rz	NR1	NR2	Description	Acre Yield Sugar Lbs	Beets Tons	Sucrose %	Beets/100' No.	RJAP %	Powdery Mildew 9/27
Checks										
Beta 4430R	√			3-21-03	17796	52.03	17.09	159	87.2	3.6
Phoenix	√			9-12-03	15960	49.53	16.13	164	86.9	4.3
Roberta				3-25-03	18092	53.06	17.02	165	86.7	4.9
P318-6H5	√		√	C833-5CMS x P118-6, (CP08)	16803	48.98	17.13	162	83.9	2.9
Syngenta hybrids										
Hil-1	√	√	√	4-22-03	14684	44.54	16.46	147	84.9	6.1
Hil-2	√	√	√	4-22-03	14137	44.51	15.91	139	85.3	5.3
Hil-3	√	√	√	4-22-03	14956	51.90	14.43	152	86.2	5.5
USDA MM breeding lines										
N312	√		√	PMR-RZM-NR N112	15100	45.40	16.61	161	84.0	1.0
N372	√		√	RZM-ER-NR N172	14218	43.48	16.39	153	83.0	2.5
Betaseed hybrids										
2VK0305	√		√	9-12-03	16279	51.60	15.80	153	85.9	4.3
OVK6280	√		√	9-12-03	15201	52.81	14.44	156	85.1	5.0
2AP0852		√	√	9-12-03	15388	45.20	17.02	168	86.7	7.6
2EN5066		√	√	9-12-03	14639	47.31	15.51	154	87.3	7.0
USDA Experimental hybrids										
1927-4H50	√		√	C790-15CMS x RZM 9927-4, (C927-4)	17030	50.03	16.98	152	88.4	4.6
P318-6H50	√		√	C790-15CMS x P118-6, (CP08)	15729	48.42	16.26	156	84.1	3.5
P207/8H50	√		√	C790-15CMS x P007/8, (CP07)	16253	47.72	17.04	164	84.8	3.0
N330-5H50	√			C790-15CMS x RZM N230-5-#(C) (g)	16579	49.43	16.76	164	84.4	3.8
N325H50	√		√	C790-15CMS x RZM N224(C) (g)	17756	51.84	17.14	157	85.5	4.8
R378H93	√		√	N265-31HO x RZM R178, (C78/3)	14650	45.20	16.20	152	86.1	4.8

TEST 704. CYST NEMATODE COMMERCIAL AND EXPERIMENTAL HYBRIDS UNDER NONDISEASED CONDITIONS, SALINAS, 2004

(cont.)

Variety	Resistance Rz	NR1	NR2	Description	Acre Yield Sugar Lbs	Beets Tons	Sucrose %	Beets/ 100' No.	RJAP %	Powdery Mildew 9/27
USDA Experimental hybrids (cont.)										
1927-4H5	√			C833-5CMS x RZM 9927-4,(C927-4)	15552	46.76	16.63	156	83.3	3.9
Y375H50		√		C790-15CMS x RZM Y275	15824	47.41	16.66	160	84.7	3.9
Y367-5H50			√	C790-15CMS x RZM Y167-5	15620	46.34	16.81	152	84.4	2.6
Y367			√	RZM-ER-% Y167,(C67/2)	14582	42.65	17.11	157	84.5	2.6
R381-22H50		√		C790-15CMS x RZM R181-22,(C81-22)	16781	49.03	17.11	160	85.8	2.6
Mean					15817.1	48.13	16.44	156.8	85.4	4.2
LSD (.05)					1388.3	3.24	0.82	9.9	1.9	1.1
C.V. (%)					8.9	6.83	5.07	6.4	2.3	25.8
F value					5.3**	7.04**	6.90**	3.4**	4.0**	16.0**

NOTES: Test 704 was grown in Block 6, Spence Field. In 2002, the field was fumigated with methyl bromide/ chloropricin and strawberries grown in 2003. Weed and disease problems appeared minimal, although at harvest, the crew reported the occurrence of mild rhizomania. At harvest, soil cores were taken and will be analyzed for sugarbeet cyst nematode and rhizomania.

Also see Test B504 from Imperial Valley and Tests 3504 and 5704 from Salinas. Test B504 was under severe SBCN/mild rzm conditions; Test 704 was under non-diseased conditions; Test 5704 was under mild rhizomania conditions; and Test 3504 was under moderate SBCN/high rhizomania conditions.

Resistance: Rz = Holly (Rz1)) or other sources of resistance to rhizomania. USDA lines and hybrids usually segregate for resistance to rhizomania. NR1 = Beta procumbens source of resistance to SBCN. N330-5H50 was misclassified and does not appear to have the B.procumbens gene, Hs. N325H50's pollinator segregates for Hs. R378H93's mm female line may be homozygous HsHs. C927-4 appears to segregate for resistance to SBCN from B.vulgaris subsp.maritima from C51. CP07 and CP08 may segregate for SBCN resistance from Bvm, WB242, as does CN12. CN72's resistance is from a different wild beet source. Y367 may have resistance to both SBCN and rhizomania from C51. Monogerm female line C790-15CMS does not have major gene resistance for rhizomania or SBCN. C833-5CMS is nearly homozygous Rz1Rz1.

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TEST 804. EVALUATION OF HYBRIDS WITH S$_1$ PROGENY LINE POLLINATORS, SALINAS, CA, 2004

Planted: March 19, 2004
Harvested: September 28, 2004

48 entries x 8 reps., RCB(e)
1-row plots, 22 ft. long

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	RJAP %
Checks						
HH142	9-12-03	16255	47.52	17.11	158	85.4
Beta 4430R	8-21-03	19537	58.65	16.70	161	86.7
Phoenix	9-12-03	18023	54.52	16.54	166	89.2
Beta 4001R	8-25-03	19123	56.18	17.02	165	86.7
Populations hybrids						
3931H50	C790-15CMS x RZM 2931, (C931)	17065	51.17	16.69	156	85.4
3941H50	x RZM 2941, (C941)	17054	50.44	16.91	158	86.5
Z325H50	x RZM Z225, (Cz25/2)	17468	51.45	16.99	160	85.5
CR311H50	x RZM CR211, (CR11)	16728	50.70	16.49	156	85.2
3942H50	x RZM 2942	15766	48.93	16.15	165	84.7
3943H50	C790-15CMS x 2943 (C)	17724	52.11	17.01	159	85.1
3941H5	C833-5CMS x RZM 2941, (C941)	16899	49.49	17.08	161	84.0
P207/8H50 Iso	C790-15CMS x RZM-PMR-NR P007/8, (CP07)	18096	54.62	16.59	165	85.3
Retests and new increases						
3927-4H50	C790-15CMS x RZM 9927-4,C927-4	18550	55.93	16.61	165	86.9
2930-19H50	x RZM 1930-19,C930-19	17745	53.11	16.70	159	84.9
Z325-9H50	x Z825-9,CZ25-9	18953	53.77	17.65	161	85.3
2936-16H50	x RZM 0936-16	16946	49.13	17.27	158	84.8
2930-35H50	C790-15CMS x RZM 1930-35,C930-35	17612	50.03	17.60	159	86.0
1929-4H50	x RZM 9929-4	16949	49.53	17.10	156	83.7
2941-20H50	x 0941-20	17264	51.95	16.63	153	84.3
2933-14H50	x 0933-14	17823	51.50	17.30	161	84.4
3931-56H50	C790-15CMS x RZM 1931-56	18563	54.62	17.00	158	86.0
Z331-14H50	x RZM Z131-14	17116	49.98	17.10	161	85.2
Roberta	3-25-03	18305	55.88	16.40	165	85.8
Angelina	3-10-03	18205	52.91	17.21	160	87.3

(cont.)

Variety	Description	Acre Yield Sugar Lbs	Beets Tons	Sucrose %	Beets/ 100' No.	RJAP %
Selected S₁ lines						
3931-120H50	C790-15CMS x 1931-120	16792	49.58	16.94	163	85.1
3941-107H50	x 1941-107	18144	52.81	17.19	163	85.3
3941-112H50	x 1941-112	18693	54.22	17.29	165	84.7
3933-107H50	x 1933-107	17727	53.46	16.60	161	84.4
3933-113H50	x 1933-113	17266	51.60	16.77	170	84.6
3933-118H50	x 1933-118	18065	53.56	16.88	153	85.9
Z325-105H50	x Z125-105	17201	49.28	17.48	156	85.4
Z325-109H50	x Z125-109	17270	50.34	17.15	152	85.3
CR311-6H50	C790-15CMS x CR111-6	16751	49.23	17.00	160	84.4
CR311-41H50	x CR111-41	17107	52.00	16.45	164	84.5
CR311-88H50	x CR111-88	17947	54.02	16.65	165	85.1
CR310-14-2H50	x RZM CR110-14-2	15660	47.01	16.69	156	83.8
03-FC1030-15H50	C790-15CMS x 01-FC1030-15	17169	48.43	17.74	168	84.5
03-FC1030-16H50	x 01-FC1030-16	17096	50.74	16.88	161	84.6
Hybrids with C833-5CMS						
1927-4H5	C833-5CMS x RZM 9927-4,C927-4	18014	52.00	17.30	159	84.1
3931-56H5	x RZM 1931-56	17463	50.47	17.31	156	82.8
Z331-14H5	C833-5CMS x RZM Z131-14	16801	46.48	18.09	153	85.0
2930-19H5	x RZM 1930-19,C930-19	17092	49.28	17.35	159	84.7
2936-16H5	x RZM 0936-16	16590	46.51	17.85	160	83.4
1929-4H5	x RZM 9929-4	17635	48.97	18.00	152	83.7
P318-6H5	C833-5CMS x P118-6, (CP08)	17920	52.15	17.19	159	83.8
2930-35H5	x RZM 1930-35	16247	45.45	17.89	157	82.9
3931H5	x RZM 2931, (C931)	17075	49.93	17.11	154	83.8
3942H5	x 2942	16428	47.52	17.31	153	83.2

TEST 804. EVALUATION OF HYBRIDS WITH S_1 PROGENY LINE POLLINATORS, SALINAS, CA, 2004

(cont.)

		Acre Yield			Beets/ 100'	RJAP
Variety	Description	Sugar Lbs	Beets Tons	Sucrose %	No.	%
Mean		17456.7	51.23	17.06	159.7	85.0
LSD (.05)		1144.0	3.23	0.65	9.1	1.8
C.V. (%)		6.7	6.41	3.88	5.8	2.1
F value		4.1**	5.95**	3.50**	1.8**	3.6**

NOTES: Multigerm,S^f,A:aa populations similar to C931, C941, CZ25/2, and CR11 have been developed for combined disease resistance and performance by population improvement methods and infusion of new germplasm. From these or other S^f,MM populations, individual plants were selfed under paper bags in the greenhouse and S_1 progeny, per se, tests run at Salinas and Brawley, CA. From about 250 progenies each year, about 24 are selected based upon sugar yield, disease resistance, nonbolting, %sugar, etc. These selected S_1 progenies are increased in isolation chambers and crossed to a common tester, usually C790-15CMS. Also, sometimes C833-5CMS is used. Two years following the S_1 progeny tests, the experimental hybrids with the selected progenies are tested at Salinas and Brawley, CA. Test 804 is one of these tests.

See Tests B204 from Imperial Valley and 5804 at Salinas. Also see 1104 and 4404 for performance of the progeny lines, per se.

Essentially no diseases or pests were observed in Test 804.

TEST 904. EVALUATION OF HYBRIDS WITH SELF-STERILE (SˢSˢ) POLLINATORS, SALINAS, 2004

48 entries x 8 reps., RCB(e)
1-row plots, 22 ft. long

Planted: March 19, 2004
Harvested: September 22, 2004

| Variety | Description | Acre Yield | | | Beets/100' | RJAP |
		Sugar Lbs	Beets Tons	Sucrose %	No.	%
Checks						
Beta 4001R	8-25-03	17457	51.41	16.96	159	85.6
Phoenix	9-12-03	16760	51.17	16.40	160	87.7
Beta 4430R	8-21-03	17835	53.93	16.54	162	85.6
HH142	9-12-03	15115	46.40	16.31	160	84.4
Line hybrids						
R378H50	C790-15CMS x RZM R178, (C78/3)	15308	45.45	16.90	160	84.1
Y391H50	x RZM-ER-% Y191	15754	47.26	16.70	163	83.7
Y392H50	x RZM Y292	15527	46.64	16.65	161	82.1
Y375H50	x RZM Y275	14827	44.35	16.74	163	84.9
R321H50	x RZM R221, (C26,C27)	15593	47.83	16.33	159	82.6
P207/8H50(Sp)	x P007/8, (CP07)	16270	49.72	16.38	157	83.4
Retests and new increases						
P318-6H50(Sp)	C790-15CMS x P118-6, (CP08)	15253	48.02	15.84	153	84.6
P318-6H50(Iso)	x PMR-RZM P118-6, (CP08)	15995	49.36	16.20	153	83.8
Y367-5H50	x RZM Y167-5	15355	47.40	16.19	157	83.9
R280/2-9H50	x R080/2-9	16336	48.07	17.00	159	84.6
Y267-21H50	x Y067-21	15882	46.21	17.23	149	83.4
R280-6H50	x R080-6	16928	51.08	16.60	156	83.5
Y269-8H50	x Y069-8	14496	43.35	16.74	172	82.9
R381-22H50	x RZM R181-22, (C81-22)	15626	46.35	16.88	166	84.3
R378-6H50	x RZM R178-6	15287	47.36	16.16	165	83.0
R380-21H50	x RZM R180-21	16344	49.03	16.69	162	84.3
Y368-8H50	C790-15CMS x RZM Y168-8	16297	48.26	16.85	157	84.7

TEST 904. EVALUATION OF HYBRIDS WITH SELF-STERILE (S^sS^s) POLLINATORS, SALINAS, 2004

(cont.)

Variety	Description	Acre Yield			Beets/ 100' No.	RJAP %
		Sugar Lbs	Beets Tons	Sucrose %		
Selected FS lines						
Y390-40H50	C790-15CMS x Y190-40	16249	47.26	17.19	172	83.1
Y390-43H50	x Y190-43	16566	48.26	17.15	169	84.0
Y390-83H50	x Y190-83	15632	45.92	17.05	169	84.7
Y390-98H50	x Y190-98	15811	48.60	16.26	166	84.9
R376-89-10H50	x R176-89-10	16328	46.45	17.60	168	83.4
R376-89-4H50	x R176-89-4	15926	47.21	16.86	166	83.7
R376-89-5-4H50	x RZM R176-89-4-5,(C76-89-5-4)	16371	49.41	16.58	168	83.7
Y375-9H50	x Y175-9	15520	46.74	16.61	168	84.6
Y375-13H50	x Y175-13	15849	47.26	16.77	165	83.0
Y375-20H50	C790-15CMS x Y175-20	15477	46.83	16.56	169	83.3
Roberta	3-25-03	16657	52.03	16.05	168	85.0
Hybrids with C833-5CMS						
R378H5	C833-5CMS x RZM R178,(C78/3)	15060	44.01	17.05	154	82.8
R381-22H5	x RZM R181-22,(C81-22)	16044	47.07	17.08	155	82.3
P318-6H5	x P118-6,(CP08)	15974	47.93	16.68	157	81.1
P207/8H5	x P007/8,(CP07)	15215	44.16	17.24	155	83.2
Y367-5H5	x RZM Y167-5	14790	42.57	17.36	148	82.0
R378-6H5	x RZM R178-6	15667	45.78	17.14	159	82.5
R380-21H5	x RZM R180-21	15376	43.63	17.63	159	83.4
Y368-8H5	x RZM Y168-8	15592	45.11	17.29	155	83.4
R280/2-9H5	C833-5CMS x %R080/2-9	14848	42.63	17.42	152	81.3
Angelina		17948	52.37	17.15	161	85.0
USDA experimental hybrids (retests)						
Y269-39H50	C790-15CMS x Y069-39	15446	45.92	16.80	160	82.4
Y267-34H50	x Y067-34	15480	46.88	16.51	164	83.7

TEST 904. EVALUATION OF HYBRIDS WITH SELF-STERILE (S^{s}S^s) POLLINATORS, SALINAS, 2004

(cont.)

| Variety | Description | Acre Yield | | | | Beets/ 100' | RJAP |
		Sugar Lbs	Beets Tons	Sucrose %		No.	%
USDA experimental hybrids (retests) (cont.)							
Y271-14H50	C790-15CMS x Y071-14	16044	49.69	16.17		166	82.8
R243-14H50	x R043-14	17158	51.80	16.61		162	84.9
Y291H5	C833-5CMS x RZM Y191	15455	44.25	17.46		153	82.8
Z325H5	x RZM Z225	14631	42.49	17.26		155	81.9
Mean		15861.6	47.31	16.79		160.7	83.7
LSD (.05)		1227.4	3.48	0.57		9.2	1.6
C.V. (%)		7.9	7.46	3.47		5.8	2.0
F value		3.1**	4.66**	4.34**		3.3**	4.4**

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TEST 1004. EVALUATION OF TOPCROSS HYBRIDS, SALINAS, CA, 2004

48 entries x 8 reps., RCB(e)
1-row plots, 22 ft. long

Planted: March 19, 2004
Harvested: September 22, 2004

Variety	Description		Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/ 100' No.	RJAP %
Checks							
Phoenix	9/12/03		15270	48.79	15.65	161	85.6
HH142	9/12/03		14020	43.44	16.14	159	83.1
Beta 4001R	8/25/03		16730	51.27	16.33	166	83.5
Beta 4430R	8/21/03		16313	49.41	16.50	158	86.3
Angelina	2003		17160	51.70	16.60	168	85.1
Roberta	3/25/03		15721	48.98	16.02	166	85.2
Topcrosses with C78							
R378H50	C790-5CMS x RZM R178	(C78/3)	14793	44.87	16.48	158	83.0
R378H5	C833-5CMS x RZM R178	(C78/3)	15225	44.87	16.96	155	82.5
R378H3	C562HO x RZM R178	(C78/3)	13359	40.62	16.41	157	82.9
R378H62	2835-8H5 x RZM R178	(C78/3)	14152	42.77	16.54	164	81.9
R378H63	2835-10H5 x RZM R178	(C78/3)	14544	43.82	16.63	163	82.2
R378H64	2835-24H5 x RZM R178	(C78/3)	14740	44.78	16.48	162	81.5
R378H66	2836-13H5 x RZM R178	(C78/3)	14408	43.92	16.41	160	82.1
R378H67	0837-6H5 x RZM R178	(C78/3)	14295	43.39	16.49	160	82.8
R378H59	2869-15H5 x RZM R178	(C78/3)	14603	44.30	16.49	161	82.4
R378H60	2840-9H5 x RZM R178	(C78/3)	15182	47.21	16.09	160	82.0
R378H68	2848-1H5 x RZM R178	(C78/3)	15325	45.83	16.70	164	82.9
R378H80	2810-17H5 x RZM R178	(C78/3)	14163	42.30	16.75	161	82.9
R378H81	2810-19H5 x RZM R178	(C78/3)	14505	43.44	16.69	157	82.0
R378H77	1833-5-8HO x RZM R178	(C78/3)	14932	42.68	17.51	151	83.7
R378H78	1833-5-11HO x RZM R178	(C78/3)	15468	45.35	17.08	153	82.2
R378H74	02-1015HO x RZM R178	(C78/3)	15082	44.92	16.79	155	83.1
R378H73	02-124HO x RZM R178	(C78/3)	14843	44.25	16.77	161	81.6
R378H42	2842HO x RZM R178	(C78/3)	14316	43.58	16.42	155	80.9

TEST 1004. EVALUATION OF TOPCROSS HYBRIDS, SALINAS, CA, 2004

(cont.)

Variety	Description		Acre Yield		Sucrose	Beets/100'	RJAP
			Sugar Lbs	Beets Tons	%	No.	%
Topcrosses with C78 (cont.)							
R378H92	2790H5	x RZM R178 (C78/3)	15148	45.83	16.52	161	82.6
R378H93	N265-31HO	x RZM R178 (C78/3)	12850	40.96	15.70	154	83.0
R378H94	N265-9HO	x RZM R178 (C78/3)	14097	43.97	16.05	161	81.6
R378H99	N265(C)HO	x RZM R178 (C78/3)	13459	42.63	15.79	159	83.2
Topcrosses with popn-931							
3931H5	C833-5CMS	x RZM 2931	15305	44.68	17.13	157	82.1
3931H74	02-FC1015HO	x RZM 2931	14327	42.85	16.73	157	80.5
3931H73	02-FC124	x RZM 2931	14700	44.97	16.38	159	80.4
3931H59	2869-15H5	x RZM 2931	14599	44.59	16.42	160	82.9
3931H62	2835-8H5	x RZM 2931	15009	45.11	16.66	167	81.7
3931H63	2835-10H5	x RZM 2931	15203	45.16	16.83	160	81.5
3931H64	2835-24H5	x RZM 2931	14896	45.40	16.48	166	81.7
3931H66	2836-13H5	x RZM 2931	14924	44.73	16.68	160	82.2
3931H60	2840-9H5	x RZM 2931	15854	48.98	16.16	162	83.1
3931H68	2848-1H5	x RZM 2931	14426	44.30	16.27	168	83.3
3931H80	2810-17H5	x RZM 2931	15214	45.68	16.64	160	81.9
3931H81	2810-19H5	x RZM 2931	14282	43.44	16.42	157	81.4
3931H42	2842HO	x RZM 2931	14917	45.49	16.40	157	82.5
3931H3	97-C562HO	x RZM 2931	13114	41.10	15.94	165	84.0
Experimental hybrids							
3942H5	2833-5NBHO	x RZM 2942	14810	44.11	16.79	161	81.7
3943H5	2833-5NBHO	x RZM 2943(C)	14721	43.73	16.84	164	81.2
Experimental hybrids with _Rz2_							
R324/5H5	2833-5NBHO	x R824, (C79-2,-3;WB41,42)	14420	43.99	16.39	143	81.1
R324H5		x R724, (C79-2; WB41)	14596	44.44	16.42	163	81.2

(cont.)

| Variety | Description | Acre Yield | | | Beets/ 100' | RJAP |
		Sugar Lbs	Beets Tons	Sucrose %	No.	%
Experimental hybrids with *Rz2* (cont.)						
R325H5	2833-5NBHO x R725, (C79-3; WB42)	13645	42.44	16.07	164	80.9
R337H5	x R637, (C79-9; WB151)	13377	40.58	16.49	157	81.4
Mean		14730.1	44.70	16.48	160.0	82.5
LSD (.05)		1096.8	3.06	0.52	9.9	1.7
C.V. (%)		7.6	6.95	3.22	6.3	2.1
F value		4.5**	4.91**	3.67**	1.8**	4.4**

TEST 204. PERFORMANCE OF HYBRIDS UNDER BYV INFECTED CONDITIONS, SALINAS, CA, 2004

24 entries x 8 reps., RCB(e)
1-row plots, 22 ft. long

Planted: March 19, 2004
Harvested: October 14, 2004
Inoculated BYV: May 13, 2004

| Variety | Description | | Acre Yield | | | Sucrose % | Beets/ 100' No. | RJAP % | Powdery Mildew 9/27 | Virus Yellows Scores | | | |
			Sugar Lbs	Loss %	Beets Tons					7/28	8/06	8/19	Mean
Checks													
Beta 4001R	8/25/03		14721	16.3	45.85	16.08	160	84.1	1.4	5.3	5.0	5.0	5.4
Phoenix	9/12/03		12171	23.6	40.21	15.15	155	86.6	3.1	3.9	4.3	5.0	4.8
HH142	9/12/03		12234	19.6	38.19	16.01	149	83.4	2.9	3.6	3.6	4.6	4.4
Beta 4430R	8/21/03		14169	18.8	43.84	16.16	158	85.9	4.1	5.5	5.1	6.3	5.9
Experimental hybrids													
Y291H5	C833-5CMS	x RZM Y191	13608	6.5	41.69	16.33	149	81.1	2.4	3.4	2.6	2.6	3.3
Y391H50	C790-15CMS	x RZM-ER-% Y191	12510	19.1	39.91	15.68	151	85.2	1.4	3.0	3.0	2.9	3.4
R376-89-4H50	C790-15CMS	x R176-89-4	13195	10.6	40.16	16.41	156	84.4	1.9	2.1	2.0	1.6	2.4
R381-22H5	C833-5CMS	x RZM R181-22	14239	3.0	42.89	16.61	146	84.0	1.5	2.4	2.1	1.6	2.5
Y375H50	C790-15CMS	x RZM Y275	13190	14.9	41.27	15.96	153	83.1	2.5	3.4	3.4	3.1	3.8
Y392H50	C790-15CMS	x RZM Y292	12887	14.5	40.71	15.84	157	82.5	2.0	2.8	3.1	2.6	3.3
Z210H5	C833-5CMS	x Z010(C)	11064	25.4	33.71	16.41	149	80.9	4.8	4.8	5.3	4.9	5.2
2930-35H5	C833-5CMS	x RZM 1930-35	12911	13.8	38.29	16.86	151	81.2	3.9	4.1	4.5	4.9	4.9
2930-19H5	C833-5CMS	x RZM 1930-19	13555	7.0	41.30	16.41	145	82.3	1.8	2.8	2.8	1.9	3.0
R380-21H5		x RZM R180-21	13584	14.8	41.81	16.20	147	81.4	3.1	3.9	3.6	3.0	4.0
Y368-8H5		x RZM Y168-8	13727	7.5	41.03	16.73	151	83.4	2.4	3.0	3.3	2.4	3.4
R378-6H5		x RZM R176-6	13769	9.7	41.27	16.69	149	83.4	3.3	4.6	4.3	4.0	4.5
R378H5	C833-5CMS	x RZM R178	14312	4.5	43.43	16.45	147	82.3	3.0	3.4	3.0	2.0	3.3
Y367-5H5		x RZM Y167-5	14081	2.2	42.33	16.64	153	83.1	1.6	3.0	2.8	2.3	3.1
P318-6H5		x P118-6	14057	10.4	43.79	16.05	148	84.0	2.1	2.4	1.5	1.1	2.3
3931-56H5		x RZM 1931-56	14268	12.2	43.59	16.38	147	82.6	1.1	2.1	1.6	0.6	2.1
3931H5		x RZM 2931,1931	13472	13.3	41.17	16.36	151	84.0	2.0	3.5	3.4	2.5	3.5
3941H5		x RZM 2941,1941	13441	11.8	40.41	16.63	155	83.7	2.1	3.8	3.1	2.5	3.5
3942H5		x RZM 2942	13179	4.3	39.60	16.64	149	83.1	1.6	3.4	2.9	2.1	3.3
Z325H5		x RZM Z225,Z125(C)	13029	12.9	39.76	16.40	145	83.5	3.1	3.9	4.1	3.1	4.1

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TEST 204. PERFORMANCE OF HYBRIDS UNDER BYV INFECTED CONDITIONS, SALINAS, CA, 2004

(cont.)

Variety Description	Acre Yield			Sucrose	Beets/100'	RJAP	Powdery Mildew	Virus Yellows Scores			
	Sugar Lbs	Loss %	Beets Tons	%	No.	%	9/27	7/28	8/06	8/19	Mean
Mean	13390.5		41.09	16.29	150.9	83.3	2.5	3.5	3.3	3.0	3.7
LSD (.05)	999.1		2.74	0.52	7.5	2.4	0.9	0.7	0.8	2.0	0.6
C.V. (%)	7.6		6.77	3.26	5.0	2.9	35.7	19.8	24.0	33.6	15.7
F value	5.3**		5.99**	4.23**	2.4**	2.8**	9.2**	13.8**	13.3**	15.8**	24.0**

NOTES: Tests 204 and 604 are companion tests. Test 204 was inoculated May 13, 2004 with *Beet yellows virus* (BYV).
%Loss is the relative sugar yield loss calculated from the corresponding means in each test. Inoculum was produced
by H.-Y.Liu and J.L.Sears. A source of BYV was passed through *chenopodium captitatum* and from plants with severe
vein clearing, transferred to sugarbeet plants used to produce viruliferous aphids for the field inoculation. BWYV
and BChV could not be detected in the source plants or subsequently from plants inoculated in the field. Little
natural VY infection appeared to occur.

Virus yellows foliar symptoms were scored on a scale of 0-9, where 9 = 90-100% of the mature leaf area yellowed.
Scores were made on 7/28, 8/06, 8/19 by DP.

At harvest, test 204 showed moderate rhizomania.

Correlations within VY inoculated test 204

	SY	RY	%S	RJAP	% loss
BYV mean	-0.30NS	-0.23NS	-0.18NS	0.12NS	0.54**
BYV 7/28	-0.12NS	-0.11NS	-0.04NS	0.07NS	0.50*
BYV 8/06	-0.34NS	-0.31NS	-0.11NS	0.05NS	0.61**
BYV 8/19	-0.36NS	-0.27NS	-0.27NS	0.20NS	0.68**
% sugar	0.32NS	-0.06NS		-0.51*	-0.63**
% loss	-0.65**	-0.43*	-0.63**	0.24NS	

Correlations between corresponding tests

	Non-inoculated test (Test 604)			
VY Inoc.	SY	RY	%S	RJAP
SY	0.31NS	0.30NS	-0.19NS	-0.13NS
RY	0.51*	0.55**	-0.45*	0.10NS
% sugar	0.43*	-0.58**	0.63**	-0.61**
RJAP	0.43*	0.60**	-0.70**	0.62**
% loss	0.53*	0.48*	-0.15NS	0.45*
BYV mean	0.48*	0.42*	-0.07NS	0.30NS

TEST 5704. CYST NEMATODE COMMERCIAL AND EXPERIMENTAL HYBRIDS UNDER RHIZOMANIA CONDITIONS, SALINAS, 2004

24 entries x 8 reps., RCB(e)
1-row plots, 22 ft. long

Planted: May 4, 2004
Harvested: October 25, 2004

Variety	Resistance Rz	NR1	NR2	Description	Acre Yield Sugar Lbs	Beets Tons	Sucrose %	Beets/100' No.	RJAP %
Checks									
Beta 4430R	✓			3-21-03	14705	44.44	16.54	221	83.1
Phoenix	✓			9-12-03	13290	40.86	16.26	216	84.4
Roberta				2-25-04	13482	43.74	15.38	200	83.6
P318-6H5	✓		✓	C833-5CMS x P118-6, (CP08)	14295	44.14	16.19	211	81.2
Syngenta hybrids									
Hil-1	✓		✓	4-22-03	12862	39.86	16.11	194	80.9
Hil-2	✓	✓	✓	4-22-03	12971	41.13	15.77	185	81.9
Hil-3	✓	✓	✓	4-22-03	10790	39.71	13.55	192	83.1
USDA MM breeding lines									
N312		✓	✓	PMR-RZM-NR N112, (CN12)	11819	37.14	15.95	210	81.6
N372	✓		✓	RZM-ER-NR N172, (CN72)	12130	39.35	15.43	195	81.8
Betaseed hybrids									
2VK0305	✓			9-12-03	13198	41.17	16.02	191	82.8
OVK6280	✓	✓		9-12-03	11545	37.64	15.30	210	82.9
2AP0852	✓		✓	9-12-03	12696	39.00	16.27	216	82.5
2EN5066		✓	✓	9-12-03	13064	42.58	15.34	218	85.7
USDA Experimental hybrids									
1927-4H50	✓	✓	✓	C790-15CMS x RZM 9927-4, (C927-4)	14626	45.10	16.20	210	82.8
P318-6H50	✓	✓	✓	C790-15CMS x P118-6, (CP08)	14022	43.89	16.00	221	81.5
P207/8H50	✓	✓	✓	C790-15CMS x P007/8, (CP07)	13776	41.62	16.54	214	82.8
Angelina	✓	✓	✓	2/25/04	12827	38.80	16.51	211	82.1
N325H50	✓	✓	✓	C790-15CMS x RZM N224(C)(g)	14237	44.04	16.16	211	82.1
R378H93	✓	✓	✓	N265-31HO x RZM R178, (C78/3)	12338	39.60	15.57	198	81.9
1927-4H5	✓		✓	C833-5CMS x RZM 9927-4, (C927-4)	14441	43.43	16.63	191	81.5
Y375H50	✓		✓	C790-15CMS x RZM Y275	13391	41.12	16.27	216	81.3
Y367-5H50	✓		✓	C790-15CMS x RZM Y167-5	13797	42.38	16.27	210	82.0

A103

TEST 5704. CYST NEMATODE COMMERCIAL AND EXPERIMENTAL HYBRIDS UNDER RHIZOMANIA CONDITIONS, SALINAS, 2004

(cont.)

Variety	Resistance			Description	Acre Yield			Beets/100'	RJAP
	Rz	NR1	NR2		Sugar Lbs	Beets Tons	Sucrose %	No.	%
USDA Experimental hybrids (cont.)									
Y367	√			RZM-ER-% Y167,(C67/2)	12628	37.64	16.79	201	82.5
R381-22H5		√		C833-5HO x RZM R181-22,(C81-22)	13918	42.12	16.52	200	81.2
Mean					13202.0	41.27	15.98	205.9	82.4
LSD (.05)					963.1	2.42	0.56	15.7	2.2
C.V. (%)					7.4	5.96	3.56	7.8	2.8
F value					8.5**	7.34**	11.18**	3.6**	1.9*

Rz = Holly (Rz1) or other sources of resistance to rhizomania
NR1 = Beta procumbens source of nematode resistance.
NR2 = Possibly other sources of nematode resistance or tolerance.

Test 5704 was grown in Block 2, Spence Field. In 2001, the field was fumigated with methyl bromide/ chloropicrin and strawberries grown in 2002. The field was fallow in 2003 until it was worked flat, rhizomania/(SBCN) soil from Block 4 broadcast, disked in, and then susceptible beet drilled in August. In late October, the solid beets were disked in. Test 5704 was planted on beds in May 2004. Weed and disease problems appeared minimal. Powdery mildew and aphids were controlled. At harvest, soil cores were taken and will be analyzed for SBCN and rhizomania.

Also see Test B504 from Imperial Valley and Tests 3504 and 704 from Salinas. Test B5404 was under severe SBCN/mild rzm conditions; Test 704 under non-diseased conditions; Test 5704 under mild rzm conditions; and 3504 under moderate SBCN/high rzm conditions.

The pollinator of N325H50 segregates for Hs from B.procumbens. The mm female of R378H93 may be homozygous HsHs. C927-4 appears to segregate for resistance to SBCN from Bvm through C51(R22). CP07 and CP08 may segregate for SBCN resistance from Bvm, WB242 accession, as does CN12. CN72 may have resistance from a different wild beet source. Y367 may have resistance to both SBCN and rhizomania from C51. Monogerm female line C790-15CMS does not have major gene resistance to rzm or SBCN. C833-5CMS is nearly homozygous Rz1Rz1.

TEST 5804. EVALUATION OF HYBRIDS WITH S_1 PROGENY LINE POLLINATORS UNDER RHIZOMANIA, SALINAS, CA, 2004

48 entries x 8 reps., RCB(e)
1-row plots, 22 ft. long

Planted: May 4, 2004
Harvested: October 25, 2004

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	RJAP %
Checks						
HH142	9-12-03	12046	36.08	16.70	207	83.3
Beta 4430R	8-21-03	13804	41.37	16.66	212	83.7
Phoenix	9-12-03	12612	38.90	16.23	210	83.6
Beta 4001R	8-25-03	13775	40.71	16.90	214	82.4
Populations hybrids						
3931H50	C790-15CMS x RZM 2931	12886	39.10	16.49	194	82.0
3941H50	x RZM 2941	12481	38.55	16.19	200	82.2
Z325H50	x RZM Z225,(CZ25)	13139	39.00	16.85	211	82.8
CR311H50	x RZM CR211	12226	37.99	16.11	206	81.0
3942H50	x RZM 2942	12767	38.75	16.46	208	81.3
3943H50	C790-15CMS x 2943(C)	12755	38.60	16.54	210	82.0
3941H5	C833-5CMS x RZM 2941	12367	37.59	16.46	205	80.7
P207/8H50(Sp)	C790-15CMS x RZM-PMR-NR P007/8,(CP07)	12601	39.00	16.16	200	81.9
Retests and new increases						
3927-4H50	C790-15CMS x RZM 9927-4,C927-4	13247	40.81	16.26	215	81.9
2930-19H50	x RZM 1930-19,C930-19	12346	37.49	16.46	202	83.8
Z325-9H50	x Z825-9,CZ25-9	13106	39.30	16.70	189	81.4
2936-16H50	C790-15CMS x RZM 0936-16	11865	34.97	16.98	209	80.7
2930-35H50	x RZM 1930-35,C930-35	12543	37.24	16.85	206	82.4
1929-4H50	x RZM 9929-4	12600	37.64	16.74	202	81.9
R381-22H5	C833-5HO x R181-22	12533	37.09	16.90	201	83.4
2933-14H50	C790-15CMS x 0933-14	13779	40.41	17.05	205	82.6
3931-56H50	C790-15CMS x RZM 1931-56	13046	39.55	16.51	203	82.5
Z331-14H50	C790-15CMS x RZM Z131-14	13085	38.65	16.92	206	81.2
Roberta	2/25/04	12813	41.22	15.53	189	83.4
Angelina	2/25/04	12397	37.09	16.71	202	81.8

(cont.)

Variety	Description		Acre Yield		Sucrose %	Beets/100' No.	RJAP %
			Sugar Lbs	Beets Tons			
Selected S$_1$ lines							
3931-120H50	C790-15CMS	x 1931-120	12163	36.78	16.54	218	81.1
3941-107H50		x 1941-107	12299	37.19	16.52	214	82.7
US H11	Susc. ck., 10/14/02		10378	35.72	14.55	197	82.2
3933-107H50		x 1933-107	12448	38.40	16.23	215	81.2
3933-113H50		x 1933-113	12999	39.62	16.41	210	82.2
3933-118H50		x 1933-118	13894	42.02	16.52	194	82.1
Z325-105H50		x Z125-105	12663	38.40	16.51	196	82.0
Z325-109H50		x Z125-109	12450	37.75	16.49	199	82.1
CR311-6H50	C790-15CMS	x CR111-6	12184	37.39	16.30	200	82.3
CR311-41H50		x CR111-41	12826	39.05	16.44	207	83.3
CR311-88H50		x CR111-88	14268	43.69	16.33	216	82.3
CR310-14-2H50		x RZM CR110-14-2	12189	38.40	15.89	200	80.5
03-FC1030-15H50		x 01-FC1030-15	12577	37.24	16.91	215	82.5
03-FC1030-16H50		x 01-FC1030-16	12083	38.60	15.67	197	80.7
Hybrids with C833-5CMS							
1927-4H5	C833-5CMS	x RZM 9927-4,C927-4	13443	40.81	16.48	199	80.6
3931-56H5		x RZM 1931-56	13312	39.86	16.71	191	82.2
Z331-14H5		x RZM Z131-14	13490	38.34	17.60	196	82.4
2930-19H5		x RZM 1930-19,C930-19	12279	37.67	16.31	196	81.7
2936-16H5		x RZM 0936-16	12126	35.17	17.25	194	80.9
1929-4H5		x RZM 9929-4	12360	35.02	17.65	195	82.5
P318-6H5		x P118-6, (CP08)	13145	40.36	16.29	211	80.8
2930-35H5		x RZM 1930-35	12126	35.04	17.31	208	80.7
3931H5		x RZM 2931	12947	38.45	16.85	197	80.3
3942H5	C833-5CMS	x 2942	12250	36.88	16.60	199	80.7

TEST 5804. EVALUATION OF HYBRIDS WITH S₁ PROGENY LINE POLLINATORS UNDER RHIZOMANIA, SALINAS, CA, 2004

(cont.)

| Variety | Description | Acre Yield | | Sucrose | Beets/ 100' | RJAP |
		Sugar Lbs	Beets Tons	%	No.	%
Mean		12702.5	38.4	16.54	203.6	82.0
LSD (.05)		895.7	2.4	0.51	13.5	1.8
C.V. (%)		7.2	6.4	3.14	6.7	2.2
F value		4.2**	4.7**	7.65**	2.5**	2.1**

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TEST 5904. EVALUATION OF HYBRIDS WITH SELF-STERILE (S^{s}S^s) POLLINATORS UNDER RHIZOMANIA, SALINAS, CA, 2004

48 entries x 8 reps., RCB(e)
1-row plots, 22 ft. long

Planted: May 4, 2004
Harvested: November 2, 2004

Variety	Description		Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	RJAP %
Checks							
Beta 4001R	8-25-03		13959	40.76	17.13	219	83.3
Phoenix	9-12-03		12414	37.49	16.56	207	83.8
Beta 4430R	8-21-03		14384	41.52	17.35	218	85.1
HH142	9-12-03		12105	36.08	16.77	196	83.0
Line hybrids							
R378H50	C790-15CMS	x RZM R178, (C78/3)	13383	38.85	17.21	203	83.6
Y391H50		x RZM-ER-% Y191	13994	40.97	17.07	220	83.3
Y392H50		x RZM Y292	13304	38.95	17.09	215	81.8
Y375H50		x RZM Y275	12771	37.64	16.98	206	82.7
R321H50		x RZM R221, (C26,C27)	13083	38.60	16.94	198	83.2
P207/8H50 (Sp)		x P007/8, (CP07)	13183	39.40	16.71	212	82.2
Retests and new increases							
P318-6H50(Sp)	C790-15CMS	x P118-6, (CP08)	13694	40.71	16.81	209	82.6
P318-6H50(Iso)		x PMR-RZM P118-6, (CP08)	13210	39.15	16.88	198	82.9
Y367-5H50	C790-15CMS	x RZM Y167-5	13970	40.97	17.05	205	83.3
R280/2-9H50	C790-15CMS	x R080/2-9	13481	39.08	17.25	199	82.1
Z210H5	C833-5HO	x Z#(C)	13181	34.22	19.26	196	82.5
R280-6H50	C790-15CMS	x R080-6	14853	42.68	17.40	201	84.0
Y269-8H50	C790-15CMS	x Y069-8	12982	37.59	17.29	207	82.6
R381-22H50		x RZM R181-22, (C81-22)	13731	40.66	16.90	206	83.5
R378-6H50		x RZM R178-6	13580	40.36	16.79	209	83.8
R380-21H50		x RZM R180-21	14001	40.46	17.31	204	83.4
Y368-8H50		x RZM Y168-8	13387	38.45	17.41	201	83.9

TEST 5904. EVALUATION OF HYBRIDS WITH SELF-STERILE (SˢSˢ) POLLINATORS UNDER RHIZOMANIA, SALINAS, CA, 2004

(cont.)

Variety	Description			Acre Yield		Sucrose	Beets/100'	RJAP
				Sugar Lbs	Beets Tons	%	No.	%
Selected FS lines								
Y390-40H50	C790-15CMS	x	Y190-40	15047	41.87	17.99	218	83.2
Y390-43H50		x	Y190-43	14109	40.18	17.55	209	82.8
Y390-83H50		x	Y190-83	13265	38.45	17.27	212	83.4
Y390-98H50		x	Y190-98	13156	39.15	16.79	209	82.4
R376-89-10H50		x	R176-89-10	13126	36.95	17.76	211	82.8
R376-89-4H50		x	R176-89-4	13394	38.34	17.45	212	83.7
R376-89-5-4H50		x	RZM R176-89-4-5,(C76-89-5-4)	14528	42.33	17.15	201	83.2
Y375-9H50		x	Y175-9	13128	38.60	17.01	208	83.2
Y375-13H50		x	Y175-13	13447	39.00	17.24	217	83.6
Y375-20H50		x	Y175-20	13010	38.40	16.94	213	83.9
Roberta	2/25/04			13697	42.58	16.05	197	84.3
Hybrids with C833-5CMS								
R378H5	C833-5CMS	x	RZM R178,(C78/3)	13505	39.45	17.11	202	81.5
R381-22H5		x	RZM R181-22,(C81-22)	14012	40.46	17.31	198	83.0
P318-6H5		x	P118-6,(CP08)	14293	41.82	17.08	206	81.5
P207/8H5		x	P007/8,(CP07)	12937	38.04	17.00	200	80.9
Y367-5H5	C833-5CMS	x	RZM Y167-5	13501	38.90	17.36	193	81.6
R378-6H5		x	RZM R178-6	13749	39.60	17.35	203	83.4
R380-21H5		x	RZM R180-21	14089	40.37	17.45	194	82.6
Y368-8H5		x	RZM Y168-8	12774	35.98	17.76	190	82.9
R280/2-9H5	C833-5CMS	x	R080/2-9	13925	38.87	17.92	199	83.0
Angelina	2/25/04			12941	37.64	17.19	197	81.8
USDA experimental hybrids (retests)								
Y269-39H50	C790-15CMS	x	Y069-39	14513	43.19	16.79	208	82.2
Y267-34H50		x	Y067-34	13137	38.24	17.19	204	84.0

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TEST 5904. EVALUATION OF HYBRIDS WITH SELF-STERILE (S^{s}S^s) POLLINATORS UNDER RHIZOMANIA, SALINAS, CA, 2004

(cont.)

| Variety | Description | Acre Yield | | | Beets/ 100' | RJAP |
		Sugar Lbs	Beets Tons	Sucrose %	No.	%
USDA experimental hybrids (retests) (cont.)						
Y271-14H50	C790-15CMS x Y071-14	13762	41.12	16.74	210	82.9
R243-14H50	x R043-14	14626	42.33	17.29	209	83.7
Y291H5	C833-5CMS x RZM Y191	13999	39.35	17.80	190	83.7
Z325H5	x RZM Z225	14087	39.89	17.66	195	81.8
Mean		13591.8	39.49	17.22	204.8	83.0
LSD (.05)		972.9	2.55	0.51	13.0	1.7
C.V. (%)		7.3	6.56	2.99	6.5	2.1
F value		3.1**	4.19**	6.82**	2.7**	1.8**

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TEST 6004. EVALUATION OF TOPCROSS HYBRIDS UNDER RHIZOMANIA, SALINAS, CA, 2004

48 entries x 8 reps., RCB(e)
1-row plots, 22 ft. long

Planted: May 4, 2004
Harvested: November 3, 2004

| Variety | Description | Acre Yield | | Sucrose | Beets/100' | RJAP |
		Sugar Lbs	Beets Tons	%	No.	%
Checks						
Phoenix	9/12/03	13175	38.40	17.16	214	85.4
HH142	9/12/03	12723	37.44	16.99	208	82.7
Beta 4001R	8/25/03	14765	42.78	17.25	222	83.1
Beta 4430R	8/21/03	14842	42.73	17.35	214	84.3
Angelina	2/25/04	12834	37.14	17.27	209	83.0
Roberta	2/25/04	13525	42.78	15.81	199	82.5
Topcrosses with C78						
R378H50	C790-5CMS x RZM R178 (C78/3)	14319	41.72	17.15	212	82.9
R378H5	C833-5CMS x RZM R178 (C78/3)	13850	39.55	17.51	194	82.1
R378H3	C562HO x RZM R178 (C78/3)	11853	34.57	17.14	197	81.6
R378H62	2835-8H5 x RZM R178 (C78/3)	12769	37.21	17.16	204	82.1
R378H63	2835-10H5 x RZM R178 (C78/3)	13155	38.19	17.23	194	80.8
R378H64	2835-24H5 x RZM R178 (C78/3)	12736	37.79	16.86	205	80.7
R378H66	2836-13H5 x RZM R178 (C78/3)	13538	39.74	17.04	191	82.1
R378H67	0837-6H5 x RZM R178 (C78/3)	13742	39.91	17.21	193	81.9
R378H59	2869-15H5 x RZM R178 (C78/3)	13669	40.71	16.77	214	81.4
R378H60	2840-9H5 x RZM R178 (C78/3)	13979	41.97	16.64	207	81.2
R378H68	2848-1H5 x RZM R178 (C78/3)	13791	39.35	17.52	206	82.3
R378H80	2810-17H5 x RZM R178 (C78/3)	13874	39.55	17.55	214	82.5
R378H81	2810-19H5 x RZM R178 (C78/3)	12557	36.68	17.11	202	80.8
R378H77	1833-5-8HO x RZM R178 (C78/3)	13674	39.16	17.45	191	81.7
R378H78	1833-5-11HO x RZM R178 (C78/3)	13893	39.08	17.77	194	82.2
R378H74	02-FC1015HO x RZM R178 (C78/3)	13522	39.00	17.31	207	81.6

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(cont.)

Variety	Description	Acre Yield			Beets/100'	RJAP
		Sugar Lbs	Beets Tons	Sucrose %	No.	%
Topcrosses with C78 (cont.)						
R378H73	02-FC124HO x RZM R178 (C78/3)	13279	38.04	17.45	212	81.7
R378H42	2842HO x RZM R178 (C78/3)	12936	37.74	17.14	201	82.5
R378H92	2790H5 x RZM R178 (C78/3)	13707	39.65	17.30	206	82.2
R378H93	N265-31HO x RZM R178 (C78/3)	12222	37.12	16.45	195	81.1
R378H94	N265-9HO x RZM R178 (C78/3)	13483	40.51	16.65	208	82.7
R378H99	N265(C)HO x RZM R178 (C78/3)	11160	33.61	16.60	205	82.0
Topcrosses with popn-931						
3931H5	C833-5CMS x RZM 2931	13714	39.76	17.24	212	81.0
3931H74	02-FC1015HO x RZM 2931	13520	38.45	17.59	210	82.2
3931H73	02-FC124 x RZM 2931	13414	38.50	17.41	206	82.8
3931H59	2869-15H5 x RZM 2931	13628	41.32	16.49	206	80.9
3931H62	2835-8H5 x RZM 2931	14068	39.91	17.61	220	82.6
3931H63	2835-10H5 x RZM 2931	13613	39.67	17.14	200	79.9
3931H64	2835-24H5 x RZM 2931	12277	36.23	16.91	206	82.1
3931H66	2836-13H5 x RZM 2931	14270	41.17	17.33	197	81.7
3931H60	2840-9H5 x RZM 2931	13764	41.37	16.64	199	81.1
3931H68	2848-1H5 x RZM 2931	13533	39.91	16.96	212	81.5
3931H80	2810-17H5 x RZM 2931	14488	41.62	17.40	204	81.9
3931H81	2810-19H5 x RZM 2931	13437	39.10	17.19	191	80.8
3931H42	2842HO x RZM 2931	13293	38.65	17.24	203	81.6
3931H3	97-C562HO x RZM 2931	12895	37.96	16.99	219	81.0
Experimental hybrids						
3942H5	2833-5NBHO x RZM 2942	13342	38.40	17.39	209	81.6
3943H5	2833-5NBHO x RZM 2943(C)	13103	37.44	17.50	210	82.1

TEST 6004. EVALUATION OF TOPCROSS HYBRIDS UNDER RHIZOMANIA, SALINAS, CA, 2004

(cont.)

| Variety | Description | Acre Yield | | Sucrose | Beets/100' | RJAP |
		Sugar Lbs	Beets Tons	%	No.	%
Experimental hybrids with *Rz2*						
2833-5NBHO						
R324/5H5	x R824, (C79-2,-3; WB41,42)	14315	41.43	17.29	181	81.8
R324H5	x R724, (C79-2; WB41)	13889	39.96	17.40	214	82.1
R325H5	x R725, (C79-3; WB42)	13413	39.44	17.02	210	80.6
R337H5	x R637, (C79-9; WB151)	13228	38.60	17.15	202	79.8
Mean		13432.8	39.19	17.14	204.7	81.9
LSD (.05)		871.2	2.18	0.51	15.1	1.9
C.V. (%)		6.6	5.64	3.00	7.5	2.4
F value		5.1**	6.32**	3.98**	2.5**	2.1**

TEST 3304. HYBRIDS WITH COMBINED RESISTANCE TO RHIZOMANIA & SBCN, SALINAS, CA, 2004, BLOCK 4

24 entries x 8 reps., RCB
1-row plots, 11 ft. long

Planted: April 26, 2004
Harvested: October 28, 2004

Variety	Resistance	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	Beets/100' No.	RJAP %
Checks									
Beta 4430R	Rz	8-21-03	9994	28.94	17.38	17.0	16.5	155	81.3
HH 142	Rz	9-12-03	9321	27.32	17.16	15.0	14.5	136	79.6
Topcrosses with C78/2 & Y191									
R378H50	Rz	C790-5CMS x RZM R178, (C78/3)	9332	27.97	16.69	16.1	15.4	147	79.8
R378H5	Rz	C833-5CMS x RZM R178	9015	26.03	17.35	15.4	14.8	140	80.2
R378H68	Rz,R22	2848-1H5 x RZM R178	9232	27.14	17.15	14.0	13.8	127	80.2
R378H80	Rz,R22	2810-17H5 x RZM R178	9867	28.29	17.48	14.0	13.8	127	81.0
R378H81	Rz,R22	2810-19H5 x RZM R178	9080	26.21	17.33	16.4	16.1	149	79.9
R378H97	Rz, Bp	N267HOg x RZM R178	8983	26.67	16.83	16.0	15.4	145	80.6
R378H99	Rz, Bp	N265(C)HOg x RZM R178	9808	30.19	16.29	14.4	13.5	131	78.9
R378H93	Rz, Bp	N265-31HOg x RZM R178	8842	27.49	16.14	15.0	13.6	136	78.4
R378H94	Rz, Bp	N265-9HOg x RZM R178	9375	29.31	16.01	16.4	15.8	149	79.6
Y391H50	Rz, R22	C790-15CMS x RZM -ER-% Y191	10736	31.99	16.81	16.5	15.8	150	80.5
Nematode resistant pollinators									
N325H50	Rz, Bp	C790-15CMS x RZM N224(C)(g)	8355	25.69	16.33	17.0	16.0	155	79.4
Hil-2	Rz, Bp	4/22/03	6630	21.24	15.71	15.6	14.1	142	80.6
3927-4H50	Rz, R22	C790-15CMS x RZM 2927-4, (C927-4)	9800	30.74	15.98	17.1	16.8	156	79.6
1927-4H5	Rz, R22	C833-5CMS x RZM 9927-4, (C927-4)	9809	29.40	16.70	13.8	14.1	125	79.2
P207/8H50(Sp)	Rz, WB242	C790-15CMS x P007/8, (CP07)	10650	32.40	16.50	16.1	16.4	147	80.4
P207/8H5	Rz, WB242	C833-5CMS x P007/8, (CP07)	9838	28.81	17.15	16.0	15.3	145	80.4
P318-6H50(Sp)	Rz,R22,WB242	C790-15CMS x P118-6, (CP08)	9240	28.24	16.36	16.3	16.3	148	80.2
P318-6H5	Rz,R22,WB242	C833-5CMS x P118-6, (CP08)	10419	30.89	16.90	15.0	15.0	136	78.7

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TEST 3304. HYBRIDS WITH COMBINED RESISTANCE TO RHIZOMANIA & SBCN, SALINAS, CA, 2004, BLOCK 4

(cont.)

Variety	Resistance	Description	Acre Yield			Sucrose	Stand Count	Harv Count	Beets/ 100'	RJAP
			Sugar Lbs	Beets Tons		%	No.	No.	No.	%
Nematode resistant pollinators (cont.)										
Y375-H50	Rz,R22	C790-15CMS x RZM Y275	10039	30.74		16.34	16.5	16.6	150	79.0
Y375-9H50	Rz,R22	C790-15CMS x Y175-9	9579	29.26		16.38	16.9	16.1	153	80.2
Y367-5H5	Rz,R22	2833-5CMS x RZM Y167-5	10535	30.97		17.04	14.3	14.0	130	81.0
Susceptible check										
USH11		susceptible check	7761	25.07		15.60	15.6	14.6	142	80.3
Mean			9426.7	28.38		16.65	15.7	15.2	142.5	80.0
LSD (.05)			1309.1	4.17		0.58	2.2	2.4	20.4	2.0
C.V. (%)			14.1	14.90		3.51	14.5	15.8	14.5	2.5
F value			3.9**	2.84**		6.94**	1.7*	1.6NS	1.7*	1.1NS

A115

24 entries x 8 reps., RCB(e)
2-row plots, 22 ft. long

Planted: April 26, 2004
Harvested: November 23, 2004

Variety	Rz	NR1	NR2	Description	Sugar Yield Sugar Lbs	Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	Beets/ 100' No.	RJAP %	DI	%R(0-4)	%R(0-5)
Checks														
Beta 4430R	✓			3-21-03	14136	37.58	18.82	75.8	75.9	172	84.3	2.9	80.1	94.9
Phoenix	✓			9-12-03	13378	37.05	18.09	68.5	68.5	156	85.0	3.8	60.3	83.5
Roberta	✓			2/25/04	9815	29.30	16.75	70.9	69.6	161	84.3	4.1	54.9	73.2
P318-6H5			✓	C833-5CMSxP118-6,(CP08)	13871	36.40	19.07	74.5	73.3	169	84.1	3.8	62.3	90.6
Syngenta hybrids														
Hil-1	✓	✓		4-22-03	11394	30.85	18.46	70.1	69.4	159	84.3	3.6	64.8	88.6
Hil-2	✓	✓		4-22-03	11878	34.11	17.47	64.1	64.1	146	83.5	3.9	57.4	81.7
Hil-3		✓		4-22-03	7177	23.95	14.98	70.5	66.6	160	81.3	5.2	31.6	50.3
USDA MM breeding lines														
N312	✓	✓		PMR-RZM-NR N112, (CN12)	11650	32.25	18.08	72.6	71.4	165	82.2	4.4	42.8	77.6
N372	✓	✓		RZM-ER-NR N172, (CN72)	11741	31.18	18.88	71.5	70.0	162	84.0	4.9	34.3	62.1
Betaseed hybrids														
2VK0305	✓	✓		9-12-03	14026	38.31	18.33	59.5	58.9	135	83.0	3.7	61.7	85.2
OVK6280	✓	✓		9-12-03	7573	22.69	16.60	72.1	69.4	164	81.8	4.9	35.5	58.0
2AP0852		✓		9-12-03	14598	39.18	18.64	73.4	74.0	167	84.1	3.4	73.1	90.3
2EN5066		✓		9-12-03	10570	32.72	16.19	73.8	73.6	168	83.4	3.8	60.0	78.9
USDA Experimental hybrids														
1927-4H50		✓		C790-15CMS x RZM 9927-4, (C927-4)	13092	35.82	18.31	76.3	76.9	173	84.5	3.9	58.6	84.8
P318-6H50		✓		C790-15CMSxP118-6, (CP08)	12251	33.49	18.29	78.1	77.9	178	83.6	4.3	48.1	81.6
P207/8H50		✓		C790-15CMSxP007/8, (CP07)	13391	35.66	18.82	75.0	75.3	170	84.7	4.0	57.4	84.5

TEST 3504. CYST NEMATODE COMMERCIAL AND EXPERIMENTAL HYBRIDS UNDER RHIZOMANIA AND NEMATODE CONDITIONS, SALINAS, CA, 2004, BLOCK 4

(cont.)

Variety	Resistance Rz	NR1	NR2	Description	Sugar Yield Sugar Lbs	Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	Beets/ 100' No.	RJAP %	DI	Rhizomania Resistance %R(0-4)	%R(0-5)
USDA Experimental hybrids (cont.)														
Angelina	√			2/25/04	13409	34.85	19.28	74.9	74.9	170	84.7	3.1	80.7	95.8
N325H50	√	√		C790-15CMS x RZM N224(C)(g)	12413	33.62	18.51	72.4	72.3	164	82.9	3.9	60.0	88.6
R378H93	√	√		N265-31HO x RZM R178,(C78/3)	12692	34.73	18.27	73.9	73.0	168	84.6	3.9	56.9	84.9
1927-4H5	√		√	C833-5CMS x RZM 9927-4,(C927-4)	14109	37.35	18.90	72.5	73.1	165	83.9	3.6	68.0	91.5
Y375H50		√		C790-15CMS x RZM Y275	12989	34.88	18.63	77.0	77.8	175	83.9	4.1	53.3	85.8
Y367-5H50		√		C790-15CMS x RZM Y167-5	12725	34.86	18.27	77.4	77.6	176	83.3	3.9	60.3	85.8
Y367		√		RZM-ER-% Y167,(C67/2)	13069	34.31	19.05	72.4	73.6	164	83.8	3.6	66.9	90.1
R381-22H5	√			C833-5HO x RZM R181-22,(C81-22)	14091	36.50	19.30	73.4	74.6	167	83.9	3.2	74.7	95.5
Mean					12334.9	33.82	18.17	72.5	72.2	164.8	83.7	3.9	58.5	82.7
LSD (.05)					967.8	2.89	0.59	5.6	5.9	12.7	2.2	0.4	9.4	8.1
C.V. (%)					8.0	7.89	3.27	7.8	8.3	7.8	2.7	10.8	16.3	10.0
F value					30.9**	18.20**	25.41**	4.2**	4.6**	4.2**	1.4NS	13.9**	15.0**	15.7**

Rz = Holly (Rz1) or other sources of resistance to rhizomania

NR1 = Beta procumbens source of nematode resistance.

NR2 = Possibly other sources of nematode resistance or tolerance.

NOTES: Test 3504 was grown in Block 4, Spence field. This field has been in multiple rzm trials and has never been fumigated with methyl bromide. Except for oat cover crops, this area was fallow in 2002 and through most of the year in 2003. In august 2003, a steckling nursery under rzm conditions was grown. Test 3504 was sown into 71cm beds in April 2004. Establishment was slower than for tests in fumigated Blocks 2 and 6. Some plant loss occurred due to a root rot, possibly Rhizoctonia. Despite irrigating every 4-5 days, the plants of most entries and particularly the rzm

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TEST 3504. CYST NEMATODE COMMERCIAL AND EXPERIMENTAL HYBRIDS UNDER RHIZOMANIA AND NEMATODE CONDITIONS, SALINAS, CA, 2004, BLOCK 4

(cont.)

Variety	Resistance			Description	Sugar Yield		Sucrose	Stand Count	Harv Count	Beets/ 100'	RJAP	Rhizomania Resistance		
	Rz	NR1	NR2		Sugar Lbs	Beets Tons	%	No.	No.	No.	%	DI	%R(0-4)	%R(0-5)

NOTES: (cont.)

susceptible entries wilted by the 3rd day and the canopy of the rzm susceptible entries turned a distinct pale yellow. Powdery mildew and leaf pests were controlled. Very late, Cercospora developed moderately on Hil-1, Hil-2, and Hil-3.

Also see Test B504 from Imperial Valley and Tests 704 and 5704 from Salinas. Test B504 was under severe SBCN/mild rzm; Test 704 under non-diseased conditions; Test 5704 under mild rzm conditions.

Eggs and larvae counts: As of 12/3/04, SBCN had been counted for all 8 reps of B4430R, Phoenix, Hil-2, 2VK0305, 2AP0852, N312 and N372 for the sampling periods, early season (6/1/04) and mid-season (9/3/04) and for 4 reps for harvest (11/5/04). The same sampling procedure was used as for B504. Eight soil cores per plot were taken 2-3 inches from the beets, 12 inches deep and composited. Eggs and larvae were counted per 100 grams soil. Analyses except for means have not been done yet because of incomplete data. To date, the means for these varieties for the three sampling periods, respectively, are: B4430R (105,314,618); Phoenix (208,544,273); Hil-2 (269,79,92); 2VK0305 (216,147,16); 2AP0852 (148,130,66); N312 (294,--,165); and N372 (288,106,210). There was a very large variability from plot to plot within each variety suggesting that the initial SBCN population was low and highly variable in this trial area.

See Test 5704 for description of varieties.

Rhizomania: On November 23, 2004, Test 3504 was hand-harvested. Roots were lifted, laid out, and scored for rhizomania, topped, bagged, washed, weighed, and run thru the sugar lab. Rhizomania was scored on a scale of 0 to 9, where 0 = very smooth roots with no evidence of disease to 9 = dead due to rzm or root rot from rzm. The level of infection appeared moderately severe. Rhizomania reaction (root score) is shown as DI (disease index), average individual root rating, and %R (% resistant). %R was calculated for roots score 0-4(%R,0-4) and 0-5(%R,0-5).

TEST 6504. EVALUATION OF HYBRIDS UNDER RHIZOMANIA & CERCOSPORA LEAF SPOT, SALINAS, CA, 2004

24 entries x 4 reps., sequential
1-row plots, 11 ft. long

Planted: May 5, 2004
Harvested: November 17, 2004

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Root Rot %	Beets/ 100' No.	RJAP %	CLS Score
Checks								
Roberta	2/25/04	14764	44.98	16.40	0.0	180	84.2	4.3
Angelina	2/25/04	13014	37.34	17.48	0.0	191	82.9	3.3
Beta 4430R	8/21/03	15948	45.83	17.38	0.0	195	84.7	4.3
Monohikari		13336	37.13	17.98	0.0	191	84.0	2.3
ACH 555	CLSR check, 3/8/02	11244	31.86	17.48	9.1	184	83.2	2.0
HM-E17	3/21/02	12913	35.19	18.33	3.6	198	85.7	2.8
R378H50	C790-15CMS x R178	15861	44.82	17.67	3.6	184	85.2	3.5
3931H50	C790-15CMS x 2931	15679	44.92	17.48	0.0	186	82.0	3.3
Z325H50	C790-15CMS x Z225	15014	42.01	17.88	0.0	180	82.5	3.3
CR311	RZM CR211, CR111(C)aa x A	13346	38.83	17.17	0.0	180	81.4	1.5
Experimental hybrids								
CR311H5	C833-5HO x CR211	14465	40.31	17.85	0.0	184	82.4	2.0
CR311H50	C790-15CMS x CR211	15308	44.13	17.35	0.0	193	82.4	3.0
CR009-1H5	C833-5HO x CR909-1	15143	42.73	17.75	0.0	175	80.3	2.0
CR009-1H50	C790-15CMS x CR909-1	14756	41.96	17.52	2.2	184	80.7	2.3
03-FC1030-15H50	C790-15CMS x 01-FC1030-15	13689	38.19	17.88	0.0	180	81.7	3.3
03-FC1030-16H50	C790-15CMS x 01-FC1030-16	13188	37.98	17.30	0.0	182	84.0	2.5
CR310-14-2H50	C790-15CMS x CR110-14-2	13183	39.04	16.85	0.0	155	80.2	2.0
CR311-6H50	C790-15CMS x CR111-6	15537	43.50	17.85	0.0	170	83.3	1.8
CR311-41H50	C790-15CMS x CR111-41	14503	42.01	17.23	0.0	198	83.3	2.3
CR311-88H50	C790-15CMS x CR111-88	16911	47.27	17.80	3.0	195	82.2	1.3
2933-14H50	C790-15CMS x 0933-14	14398	40.33	17.85	0.0	191	83.4	1.5
3933-107H50	C790-15CMS x 1933-107	14554	40.74	17.85	0.0	182	82.3	2.8

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TEST 6504. EVALUATION OF HYBRIDS UNDER RHIZOMANIA & CERCOSPORA LEAF SPOT, SALINAS, CA, 2004

(cont.)

Variety	Description	Acre Yield Sugar Lbs	Beets Tons	Sucrose %	Root Rot %	Beets/ 100' No.	RJAP %	CLS Score
Experimental hybrids (cont.)								
3933-113H50	C790-15CMS x 1933-113	15632	44.56	17.55	0.0	186	83.1	1.8
3933-118H50	C790-15CMS x 1933-118	16498	46.47	17.73	2.3	180	82.8	1.3
Mean		14536.8	41.3	17.6	1.0	184.3	82.8	2.5
LSD (.05)		2457.2	6.7	0.9	6.3	25.2	3.1	1.4
C.V. (%)		12.0	11.4	3.4	451.9	9.7	2.6	39.5
F value		2.4**	2.7**	1.8*	0.9NS	1.2NS	1.7NS	3.1**

A120

TEST 5504. CBGA SALINAS CODED RHIZOMANIA TEST, SALINAS, CA, 2004

Planted: May 4, 2004
Harvested: November 1 & 12, 2004

96 entries x 8 reps., RCB
1-row plots, 22 ft. long

Code No.	Variety	Source	Sugar Yield		Sucrose %	Stand Count No.	Harv Count No.	Beets/100' No.	RJAP %	Rhizomania Resistance		Canopy (Foliar) Score
			Sugar Lbs	Beets Tons						DI	%R(0-4)	
CBGA Coded Entries												
1	241	Crystal	14037	42.62	16.45	46.4	49.5	211	83.7	1.5	98.9	1.6
2	04HX428	Holly	13501	38.13	17.70	42.1	43.5	191	81.2	2.2	97.7	1.4
3	04HX403	Holly	12681	36.90	17.19	45.3	46.8	206	82.7	1.6	98.4	2.6
4	BETA 4014R	Betaseed	15190	44.38	17.13	49.6	52.0	226	82.8	1.4	99.5	2.1
5	04HX413	Holly	13662	40.94	16.68	47.4	48.8	215	84.1	1.4	100.0	1.6
6	2J5453	Betaseed	15408	43.03	17.91	46.3	51.5	210	81.8	3.0	80.6	1.0
7	04HX425	Holly	13574	41.75	16.25	46.6	49.3	212	82.0	1.8	99.4	1.1
8	01HX004	Holly	13540	43.23	15.66	46.3	47.5	210	83.7	0.9	100.0	1.5
9	04HX417	Holly	11904	33.17	17.92	44.6	46.0	203	82.5	2.0	98.9	1.1
10	02HX219	Holly	13333	37.14	17.96	41.5	42.0	189	82.0	1.8	97.7	2.4
11	362	Crystal	15235	43.05	17.69	48.0	47.3	218	84.4	1.4	99.5	1.6
12	04HX429	Holly	12915	37.17	17.42	39.3	41.3	178	81.5	2.3	98.9	1.1
13	04HX426	Holly	13773	39.82	17.28	47.5	48.0	216	81.8	1.5	98.4	1.4
14	SS-NB7R	Holly	12894	38.22	16.87	45.0	44.8	205	81.9	1.9	96.5	2.1
15	04HX415	Holly	12640	35.44	17.85	45.6	42.5	207	83.1	2.0	96.2	2.6
16	03HX308	Holly	15188	44.13	17.23	48.0	48.8	218	81.9	1.3	100.0	1.0
17	2GK6080	Betaseed	14177	41.81	16.94	47.4	48.3	215	83.1	1.7	99.5	2.0
18	3AT064	Betaseed	15006	43.06	17.40	47.6	48.5	216	81.6	2.6	94.6	1.3
19	04HX421	Holly	13249	38.47	17.21	46.4	46.5	211	81.7	1.5	99.5	1.1
20	1GK0055	Betaseed	15258	44.33	17.23	49.6	47.0	226	84.1	1.9	99.5	2.3
21	BETA 4776R	Betaseed	14302	41.61	17.18	51.3	49.0	233	82.6	1.3	99.5	1.4
22	HH-145	Holly	12440	35.44	17.56	42.8	41.0	194	81.2	1.7	98.8	3.4
23	04HX407	Holly	13364	42.32	15.82	40.3	38.8	183	84.6	1.7	96.3	2.8
24	04HX408	Holly	12725	37.23	17.11	43.3	45.0	197	83.2	1.6	99.4	2.4
25	BETA 4440R	Betaseed	13671	41.43	16.52	47.3	48.0	215	83.0	1.5	99.5	1.9
26	1J5155	Betaseed	15105	43.79	17.24	46.8	48.3	212	84.0	2.3	96.9	1.4

(cont.)

Code No.	Variety	Source	Sugar Yield Sugar Lbs	Sugar Yield Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	Beets/ 100' No.	RJAP %	Rhizomania Resistance DI	Rhizomania Resistance %R(0-4)	Canopy (Foliar) Score
CBGA Coded Entries (cont.)												
27	4YK0903	Betaseed	14486	44.03	16.47	47.6	50.3	216	82.7	1.6	99.5	1.8
28	FALCON	Holly	12607	36.07	17.46	48.8	51.0	222	83.6	1.3	100.0	1.3
29	EAGLE	Holly	13137	39.42	16.63	46.8	49.3	212	82.0	1.6	100.0	1.8
30	04HX406	Holly	14095	41.63	16.92	43.9	45.8	199	84.2	2.3	95.3	2.6
31	1J0229	Betaseed	14093	39.24	17.97	46.8	47.0	212	82.9	1.0	99.5	1.1
32	RAPTOR	Holly	13381	40.94	16.36	44.8	45.5	203	84.0	1.4	100.0	2.9
33	04HX409	Holly	14078	41.57	16.92	44.6	45.5	203	82.8	1.7	100.0	1.4
34	04HX416	Holly	12443	36.21	17.16	45.4	46.5	206	82.0	1.7	99.4	2.4
35	4YK0902	Betaseed	14208	41.17	17.28	47.4	47.0	215	82.7	1.7	99.5	2.3
36	03HX314	Holly	12242	34.20	17.92	43.9	45.3	199	82.6	1.7	100.0	2.3
37	9J0274	Betaseed	15401	43.63	17.67	50.1	51.0	228	83.2	0.9	99.5	2.8
38	BETA 4430R	Betaseed	14878	42.69	17.43	47.4	48.5	215	83.4	0.6	100.0	1.8
39	3AT0439	Betaseed	14444	42.44	17.02	48.9	48.8	222	82.4	1.6	96.4	1.4
40	BETA 4001R	Betaseed	14351	40.78	17.62	50.4	52.0	229	83.0	1.4	100.0	3.0
41	03HX307	Holly	13960	43.69	16.01	48.5	48.0	220	81.3	1.4	98.5	1.3
42	1GK7429	Betaseed	13412	40.39	16.62	49.1	50.8	223	82.9	1.9	97.7	1.8
43	9J0158	Betaseed	12864	37.58	17.14	51.1	52.0	232	81.6	2.2	94.8	3.3
44	04HX410	Holly	11991	36.77	16.30	45.8	45.0	208	82.9	1.6	99.5	2.6
45	04HX419	Holly	15278	44.76	17.04	48.4	50.5	220	82.6	1.3	99.5	1.0
46	1J0263	Betaseed	15495	45.45	17.02	47.8	52.3	217	83.6	1.2	99.5	1.9
47	04HX427	Holly	12732	36.09	17.65	44.3	42.0	201	81.9	2.2	95.0	1.3
48	04HX404	Holly	15725	45.65	17.24	46.6	47.0	212	83.5	1.1	100.0	2.3
49	04HX401	Holly	13171	39.38	16.69	49.0	48.0	223	83.7	1.3	99.5	1.9
50	BETA 4054R	Betaseed	15997	46.86	17.06	48.6	47.8	221	81.9	1.2	100.0	1.9
51	2GK6100	Betaseed	11740	34.70	16.95	49.9	50.3	227	81.8	2.2	99.0	1.6
52	349	Crystal	15319	44.20	17.38	51.0	50.3	232	82.9	1.2	99.6	1.8

(cont.)

Code No.	Variety	Source	Sugar Yield Sugar Lbs	Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	Beets/ 100' No.	RJAP %	Rhizomania Resistance DI	%R(0-4)	Canopy (Foliar) Score
CBGA Coded Entries (cont.)												
53	2GK6113	Betaseed	14637	43.39	16.92	51.8	53.0	235	82.4	1.1	100.0	1.6
54	ALPINE	Holly	13892	42.19	16.44	49.5	52.3	225	83.7	1.5	97.7	2.4
55	444	Crystal	14553	41.12	17.71	48.9	47.0	222	82.7	2.1	97.9	1.9
56	ACCLAIM	Holly	12851	36.04	17.81	49.5	47.5	225	84.1	1.4	99.5	1.1
57	02HX204	Holly	13779	39.61	17.39	47.9	47.5	218	83.8	1.6	100.0	2.8
58	03HX313	Holly	14590	40.86	17.88	48.4	48.0	220	82.2	2.2	99.5	1.5
59	4YK0901	Betaseed	14825	41.00	18.11	47.6	48.0	216	84.4	0.9	100.0	1.4
60	04HX405	Holly	12495	38.48	16.21	30.1	33.0	137	83.9	1.5	99.1	2.3
61	3YK0801	Betaseed	15828	45.84	17.27	46.1	47.0	210	84.1	1.3	99.5	1.9
62	04HX402	Holly	13321	36.86	18.04	46.4	48.3	211	81.6	2.4	92.2	3.0
63	1GK0062	Betaseed	15911	45.33	17.57	49.0	47.3	223	83.5	1.4	100.0	2.3
64	1J0303	Betaseed	14521	41.30	17.58	47.5	47.8	216	82.3	1.2	100.0	1.9
65	3YK0802	Betaseed	15338	42.08	18.20	48.0	51.0	218	84.6	0.9	100.0	1.3
66	PHOENIX	Holly Hyb	13570	40.30	16.82	47.4	48.0	215	83.5	1.5	100.0	1.5
67	03HX309	Holly	13530	41.42	16.32	45.4	47.8	206	83.2	1.5	99.4	1.4
68	HH-142	Holly	12621	36.85	17.11	47.1	47.0	214	82.0	1.4	100.0	1.1
69	04HX420	Holly	14856	45.11	16.49	45.6	46.8	207	82.0	1.9	98.4	2.0
70	04HX418	Holly	12760	35.42	18.04	47.8	48.8	217	81.4	2.7	86.1	1.5
71	USH-11	Check	10422	33.72	15.49	48.4	47.0	220	82.0	1.5	97.2	4.3
USDA entries & checks		Male description										
72	2930-35H5	C930-35	12607	35.46	17.81	44.3	45.5	201	82.0	1.9	99.5	1.5
73	R280/2-9H5	R180/2-9	13824	39.26	17.64	43.8	42.5	199	81.2	2.1	98.3	1.1
74	US H11	Susc. check	10698	35.32	15.21	45.1	48.0	205	82.3	1.5	99.4	3.8
75	Roberta		13121	41.27	15.91	45.3	43.0	206	83.3	1.3	99.3	3.8
76	Angelina		12738	37.08	17.23	46.4	46.0	211	82.4	1.2	100.0	1.1
77	Rizor		13597	37.70	18.05	48.8	49.5	222	82.7	1.9	99.5	2.1
78	P318-6H5	CP08	14269	41.81	17.09	46.6	46.0	212	80.4	2.4	89.3	1.0

A123

TEST 5504. CBGA SALINAS CODED RHIZOMANIA TEST, SALINAS, CA, 2004

(cont.)

Code No.	Variety	Source	Male description	Sugar Yield Sugar Lbs	Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	Beets/ 100' No.	RJAP %	Rhizomania Resistance DI	%R(0-4)	Canopy (Foliar) Score
USDA entries & checks			(cont.)										
79	P207/8H5	CP07		13636	38.98	17.50	43.1	41.0	196	82.0	2.3	92.1	1.1
80	R380-21H5	R180-21		14575	41.90	17.39	48.6	46.5	221	81.1	1.4	100.0	1.0
81	Y367-5H5	R167-5		14871	42.44	17.53	45.9	45.0	209	81.5	2.3	94.2	1.0
82	Z210H5	Polish %S		12781	33.87	18.88	44.1	46.0	201	81.1	2.2	95.8	2.4
83	Z210H50	Polish %S		12283	34.70	17.69	45.9	44.3	209	81.5	2.3	96.1	3.4
84	US H11	Susc. check		10456	33.61	15.58	48.8	47.3	222	82.1	1.8	94.6	3.1
85	Rizor			13615	39.18	17.41	49.3	51.0	224	81.1	2.1	96.7	1.5
86	Angelina			13236	38.79	17.06	41.8	40.8	190	82.0	1.2	97.5	1.0
87	Roberta			13154	41.85	15.69	43.4	41.0	197	82.7	1.6	94.8	3.6
88	R381-22H5	C81-22		14400	41.93	17.21	45.3	45.3	206	81.6	1.6	100.0	1.4
89	Y368-8H5	Y168-8		13188	38.14	17.31	43.6	45.8	198	80.3	2.1	99.4	1.4
90	R378-6H5	R178-6		14005	40.17	17.44	44.0	43.5	200	81.7	1.8	98.2	1.0
91	3931-56H5	C931		14294	40.87	17.52	44.4	42.3	202	81.7	1.4	99.5	1.0
92	Z331-14H5	Z131-14		13431	37.19	18.08	43.4	41.5	197	80.9	2.0	99.5	1.9
93	Y391H50	Y291		14309	40.11	17.83	45.0	44.8	205	83.3	2.0	96.1	1.9
94	Y392H50	Y292		13770	39.75	17.34	47.4	47.8	215	82.5	2.3	94.5	1.8
95	Y375H50	Y275		12815	37.78	16.98	47.9	49.0	218	81.5	2.3	96.0	2.1
96	Y375-9H50	Y175-9		13785	40.72	16.94	44.8	48.8	203	82.5	2.2	98.0	1.1
Mean				13729.8	40.05	17.16	46.4	46.9	211.0	82.6	1.7	98.0	1.9
LSD (.05)				1103.3	3.03	0.57	3.8	5.0	17.3	1.7	0.6	6.8	0.6
C.V. (%)				8.2	7.71	3.41	8.3	7.7	8.3	2.1	26.8	5.0	34.7
F value				8.5**	9.00**	10.63**	4.7**	3.4**	4.7**	2.7**	4.0**	1.5*	10.6**

A124

(cont.)

Code No.	Variety	Source	Sugar Yield		Sucrose %	Stand Count No.	Harv Count No.	Beets/ 100' No.	RJAP %	Rhizomania Resistance		Canopy (Foliar) Score
			Sugar Lbs	Beets Tons						DI	%R(0-4)	

Foliar scores: 1 = all normal green; 2 = + 25% yellowish; 3 = 50% yellowing like Rzm : 50% green; 4 = + 75% yellowish; 5 = 100% yellowish like rzm.

In reality, the yellowing scores were for yellowing in general, not specifically for the yellowing caused by rzm, but may largely have reflected reaction to rzm except in genotypes that are naturally yellowish.

Coefficients of correlation (r) were calculated:

	RY	Foliar Score	%S
Sugar Yield	0.88**	-0.17**	0.31**
Root Yield		-0.11**	-0.16**
Foliar Score			-0.16**

The coded Rhizomania tests were run in Block 2 at Spence field. This area in the 1990's was one of the best locations for rhizomania tests, but became severely infested with *Sclerotium rolfsii*. Four years ago it was fumigated with methylbromide to eliminate *Sclerotium*. In 2002, strawberries were grown. In 2003, it remained fallow until August when it was prepared for future rzm tests. Rhizomania soil was broadcast over the area and disked in. A susceptible sugarbeet variety was then drilled in and grown four months, then disked in. In April 2004, rhizomania tests were planted. However, it is now apparent that the level of rzm was low at planting time and early infection with rzm did not occur at a high frequency, leading to poor symptom development. The coded tests were divided into 4 replication sections and planted at slightly different locations in the field, trying to hit an ideal location. Only the replications identified as 1 thru 4 were hand-harvested and scored for root symptoms. Reps 5 thru 8 were machine harvested. Analyses are presented for all 8 reps (Test 5504); reps 1-4 (Test 5504-1); and reps 5-8 (Test 5504-2).

Rhizomania was not very severe in this test. Root scores for %R (%resistant) and DI (disease index) are not reliable. The DI is too low for the susceptible checks and the %R too high.

TEST 5504. CBGA SALINAS CODED RHIZOMANIA TEST, SALINAS, CA, 2004

(cont.)

Code No.	Variety	Source	Sugar Yield		Sucrose %	Stand Count No.	Harv Count No.	Beets/ 100' No.	RJAP %	Rhizomania Resistance		Canopy (Foliar) Score
			Sugar Lbs	Beets Tons						DI	%R(0-4)	

Essentially, no other diseases or pests were observed in this trial. Powdery mildew was controlled. The agronomics appeared to be very good and uniform. For both hand-harvest and machine-harvest, two sugar samples were taken per plot. The growth throughout the season remained robust. Solid set sprinklers were not run through the test rows. Test 5504-2 was machine-harvested under fairly wet conditions following 3" of rain. Hand-harvest was delayed until the soil was dried and more easily removed from the roots to facilitate scoring.

RJAP = raw juice apparent purity = (%sucrose/%soluble solids)100.

Rhizomania scores: Only reps 1-4 (Test 5504-1) were hand-harvested and scored. After being lifted, the roots were hand shaken to remove soil and laid out. Each individual root was scored on a scale of 0 to 9, where 9 is most severe. Roots scored 0 to 4 were considered resistant and 5 to 9 were susceptible. Following scoring, two sub-samples of roots per plot were taken for sugar analyses. All roots were weighed. The samples were washed and run through the sugar lab.

Entries: Entries 1-71 were submitted by CBGA. Entries 72-96 were from USDA. For USDA entries, H5 designation means that the female parent was C833-5CMS (mm,Rz_,CMS) H50 = C790-68CMS x C790-15 (F_1CMS,rzrz,mm). Entries 72-96 were from USDA. For USDA entries, H5 designation means C790-15CMS = C790-68CMS x C790-15 (F_1CMS,rzrz,mm). The male parent is listed under the description.

A126

48 entries x 8 reps., RCB
1-row plots, 22 ft. long

Planted: May 4, 2004
Harvested: November 4 & 15, 2004

Variety	Description	Sugar Yield Sugar lbs	Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	Beets/100' No.	RJAP %	DI	Rhizomania Resistance %R(0-4)	%R(0-5)	Canopy (Foliar) Score
Checks & Calif. Comm. Hybrids												
Beta 4430R	Resist. ck., 8/21/03	16436	46.95	17.55	51.4	50.3	233	84.3	1.3	98.0	100.0	2.3
HH142	Resist. ck., 9/12/03	13474	39.05	17.27	50.1	51.8	228	82.9	2.6	85.2	99.6	1.1
Rizor	Resist. ck., 3/29/01	14037	38.57	18.22	49.6	50.0	226	83.1	3.6	61.1	94.5	1.4
Roberta	Susc. ck., 2/25/04 (pell.)	14855	44.76	16.63	48.5	47.0	220	83.3	2.3	87.9	96.4	2.8
US H11	Susc. ck., 10/4/02	11216	35.11	15.98	50.9	50.3	231	83.7	2.4	86.9	95.5	3.0
Angelina	Resist. ck., 2/25/04 (pell.)	14346	39.72	18.03	47.6	47.0	216	83.9	2.1	92.4	99.4	1.4
Beta 4001R	Resist. ck., 8/25/03	15713	44.10	17.88	51.6	50.0	235	83.4	2.6	84.8	96.5	2.0
Phoenix	Resist. ck., 9/12/03	14501	42.34	17.13	47.9	44.0	218	84.1	2.5	86.3	97.6	1.6
SMBSC Entries												
SMBSC-601	4/12/04, pelletted, blue	14795	39.83	18.59	45.0	44.5	205	83.0	4.4	32.7	84.1	1.6
-602	4/12/04, pelletted, blue	14858	40.63	18.33	44.4	43.3	202	82.6	3.0	79.0	97.1	1.1
-603	4/12/04, pelletted, green	15722	41.09	19.15	45.6	43.5	207	82.7	1.7	97.3	98.9	1.0
-604	4/12/04, pelletted, blue	14258	37.68	18.95	45.4	44.5	206	83.3	2.7	81.2	97.2	3.1
Monohikari	Susc. ck., 1/21/03	13069	36.03	18.14	48.9	47.8	222	84.7	3.2	70.3	92.7	3.0
Western Sugar Entries												
WS-1	Beta 3YK0041	13818	37.96	18.22	50.8	49.8	231	83.1	3.0	78.0	96.5	2.3
WS-2	HM 2989RZ	15582	41.87	18.65	47.9	49.3	218	82.3	2.9	80.5	96.8	1.1
WS-3	SX 0123	13857	38.88	17.82	44.3	47.3	201	83.7	3.8	53.8	91.9	2.6
WS-4	Beta 3YK0040	14812	41.32	17.97	44.8	48.3	203	82.4	3.5	65.9	97.8	1.4
WS-5	Beta 4635R	14808	42.74	17.34	45.9	49.0	209	82.3	2.8	81.6	98.5	1.0
WS-6	HH Acclaim	14017	42.05	16.68	49.1	48.0	223	83.4	2.6	84.4	99.0	1.9
WS-7	SX 230	14585	38.50	18.95	45.1	45.0	205	83.4	3.3	65.5	93.9	2.0
WS-8	SX 231	12543	33.74	18.62	46.1	48.0	210	83.6	3.6	58.8	96.9	2.6
WS-9	Monohikari (susc. ck.)	12807	35.41	18.10	46.1	48.5	210	84.8	3.1	71.7	95.4	2.8

A127

TEST 5604. WESTERN SUGAR, MICHIGAN INDUSTRY, SOUTHERN MINN.BSC, & USDA HYBRID EVALUATION UNDER RHIZOMANIA, SALINAS, CA, 2004

(cont.)

Variety	Description	Sugar Yield Sugar Lbs	Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	Beets/100' No.	RJAP %	DI	Rhizomania Resistance %R(0-4)	%R(0-5)	Canopy (Foliar) Score
Michigan Sugar Industry Entries												
HM-E17	Susc., ck., 3/21/02	11810	33.13	17.84	45.4	47.3	206	83.6	4.1	46.8	87.1	3.6
Angelina	Resist. ck., 3/25/04 (pell.)	13310	38.21	17.45	44.6	46.3	203	82.6	2.0	94.1	98.9	1.3
MS - 1	4/2/04, film (small), green	14690	39.64	18.58	44.9	47.0	204	82.6	3.5	59.1	93.7	1.3
- 2	4/2/04, film(sm),bright blue	13129	34.69	18.93	44.4	47.8	202	84.2	3.6	60.6	94.0	1.1
- 3	4/2/04, film, blue	13093	34.33	19.09	42.9	45.3	195	82.6	3.0	75.2	97.8	1.6
- 4	4/2/04, processed, brown	15638	41.78	18.74	47.0	45.8	214	83.8	3.4	63.1	90.1	1.1
- 5	4/2/04, processed, l. orange	13106	33.86	19.36	45.9	47.3	209	82.3	3.4	67.7	97.2	2.4
- 6	4/2/04, film, blue	14556	39.10	18.67	45.1	46.3	205	83.4	3.2	70.0	96.4	1.4
- 7	4/2/04, film (small), pink	12702	34.46	18.50	49.1	48.0	223	83.6	3.9	51.4	90.9	2.9
- 8	4/2/04, processed, l. orange	14370	38.39	18.73	48.0	48.3	218	83.6	3.6	64.6	89.1	1.6
- 9	4/2/04, processed, l. orange	15481	41.37	18.76	48.4	49.5	220	83.0	2.9	77.4	98.4	1.0
-10	4/2/04, film, green	14993	39.39	19.04	51.0	50.3	232	83.3	3.9	59.8	92.6	1.1
-11	4/2/04, processed, brown	15033	41.55	18.13	51.3	51.5	233	83.4	3.0	80.0	96.1	1.0
-12	4/2/04, processed, brown	13983	37.37	18.74	51.8	52.0	235	84.8	4.1	48.0	90.8	1.4
-13	4/2/04, film, blue	14215	36.30	19.57	46.9	49.0	213	82.4	2.5	92.3	99.5	2.8
-14	4/2/04, processed, brown	15329	44.32	17.31	49.5	49.3	225	83.1	2.1	92.3	99.5	1.1
-15	4/2/04, film, blue	14456	38.75	18.66	46.3	47.5	210	82.7	3.6	65.1	94.1	1.0
-16	4/2/04, processed, orange	14712	40.02	18.37	47.5	49.0	216	81.9	3.3	69.0	92.6	1.0
-17	4/2/04, film(sm),bright blue	15214	41.79	18.24	47.1	50.3	214	82.3	3.3	68.5	93.6	1.0
-18	4/2/04, film, blue	14810	38.50	19.26	44.6	46.8	203	83.0	2.5	83.1	100.0	1.4
-19	4/2/04, processed, brown	13242	34.15	19.43	46.8	49.8	212	82.9	3.5	64.7	96.7	2.0
-20	4/2/04, film (small), green	12115	33.52	18.09	47.4	47.8	215	83.6	4.3	41.2	83.9	3.5
USDA Entries												
Roberta	Susc. ck., 2/25/04 (pell.)	14303	42.90	16.69	45.9	47.0	209	83.9	2.6	82.4	93.5	3.3
R380-21H5	C833-5HO x RZM R180-21	14425	40.71	17.75	47.1	49.5	214	82.2	3.1	71.9	97.4	1.0
Y381-22H5	C833-5HO x RZM R181-22	15103	42.08	17.98	47.1	47.5	214	82.8	3.1	70.2	96.8	1.1
P318-6H5	C833-5HO x P118-6	14899	42.03	17.74	47.4	51.8	215	82.0	3.4	67.0	94.5	1.0

TEST 5604. WESTERN SUGAR, MICHIGAN INDUSTRY, SOUTHERN MINN.BSC, & USDA HYBRID EVALUATION UNDER RHIZOMANIA, SALINAS, CA, 2004

(cont.)

Variety Description	Sugar Yield Sugar Lbs	Beets Tons	Sucrose %	Stand Count No.	Harv Count No.	Beets/100' No.	RJAP %	DI	Rhizomania Resistance %R(0-4)	%R(0-5)	Canopy (Foliar) Score
Mean	14225.6	39.18	18.21	47.3	48.0	214.9	83.2	3.1	72.3	95.2	1.8
LSD (.05)	1004.2	2.69	0.54	4.1	4.8	18.8	1.6	0.8	20.4	7.8	0.7
C.V. (%)	7.2	6.98	3.03	8.9	7.1	8.9	2.0	17.4	20.2	5.8	37.0
F value	9.3**	11.98**	16.65**	2.4**	1.6*	2.4**	1.6*	6.3**	4.2**	1.9**	11.8**

Foliar scores: 1 = all normal green; 2 = + 25% yellowish; 3 = 50% yellowing like Rzm : 50% green;
4 = + 75% yellowish; 5 = 100% yellowish like rzm.

In reality, the yellowing scores were for yellowing in general, not specifically for the yellowing caused by rzm,
but may largely have reflected reaction to rzm except in genotypes that are naturally yellowish.

Coefficients of correlation (r) were calculated:

	RY	Foliar Score	%S
Sugar Yield		-0.34**	0.10NS
Root Yield	0.86**	-0.24**	-0.42**
Foliar Score			-0.14*

The coded Rhizomania tests were run in Block 2 at Spence field. This area in the 1990's was one of the best
locations for rhizomania tests, but became severely infested with *Sclerotium rolfsii*. Four years ago it was
fumigated with methylbromide to eliminate *Sclerotium*. In 2002, strawberries were grown. In 2003, it remained
fallow until August when it was prepared for future rzm tests. Rhizomania soil was broadcast over the area and
disked in. A susceptible sugarbeet variety was then drilled in and grown four months, then disked in. In April
2004, rhizomania tests were planted. However, it is now apparent that the level of rzm was low at planting time

A129

TEST 5604. WESTERN SUGAR, MICHIGAN INDUSTRY, SOUTHERN MINN.BSC, & USDA HYBRID EVALUATION UNDER RHIZOMANIA,
SALINAS, CA, 2004

(cont.)

	Sugar Yield			Stand Harv Beets/			RJAP	Rhizomania Resistance			Canopy (Foliar)
	Sugar	Beets	Sucrose	Count Count 100'			%			%R(0-5)	Score
Variety Description	Lbs	Tons	%	No.	No.	No.		DI	%R(0-4)		

and early infection with rzm did not occur at a high frequency, leading to poor symptom development. The coded tests were divided into 4 replication sections and planted at slightly different locations in the field, trying to hit an ideal location. Only the replications identified as 1 thru 4 were hand-harvested and scored for root symptoms. Reps 5 thru 8 were machine harvested. Analyses are presented for all 8 reps (Test 5604); reps 1-4 (Test 5604-1); and reps 5-8 (Test 5604-2).

Rhizomania was not very severe in this test. Root scores for %R (%resistant) and DI (disease index) are not reliable. The DI is too low for the susceptible checks and the %R too high.

Essentially, no other diseases or pests were observed in this trial. Powdery mildew was controlled. The agronomics appeared to be very good and uniform. For both hand-harvest and machine-harvest, two sugar samples were taken per plot. The growth throughout the season remained robust. Solid set sprinklers were not run through the test rows. Test 5604-2 was machine-harvested under fairly wet conditions following 3" of rain. Hand-harvest was delayed until the soil was dried and more easily removed from the roots to facilitate scoring for Test 5604-2.

RJAP = raw juice apparent purity = (%sucrose/%soluble solids)100.

Rhizomania scores: Only reps 1-4 (Test 5604-1) were hand-harvested and scored. After being lifted, the roots were hand shaken to remove soil and laid out. Each individual root was scored on a scale of 0 to 9, where 9 is most severe. Roots scored 0 to 4 were considered resistant and 5 to 9 were susceptible. Following scoring, two sub-samples of roots per plot were taken for sugar analyses. All roots were weighed. The samples were washed and run through the sugar lab.

Analyses were also run where scores 0-5 were considered resistant (%R(0-5)). By the time this test was scored, it appeared grade creep had occurred and part of 5's were actually not fully susceptible. What these disease ratings appeared to represent are combinations of rzm and other factors that influence root smoothness or roughness. No nematodes were observed in this test, so much of this may be varietal differences in smoothness.

A130

TEST B104. EVALUATION OF TOPCROSS HYBRIDS, IMPERIAL VALLEY, CA, 2003-2004

24 entries x 8 reps., RCB(E)
1-row plots, 18 ft. long

Planted: September 15, 2003
Harvested: May 19, 2004

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	Bolters %	Clean Beets %	NO3-N ppm	score
Checks									
Beta 4430R	8-21-03	13078	45.00	14.49	161	0.0	89.5	238	5.3
Phoenix	9-12-03	11705	44.26	13.25	158	0.0	94.1	279	5.4
Topcrosses with C78									
R378H50	C790-15CMS x RZM R178 (C78/3)	10238	37.13	13.83	164	3.0	92.7	235	5.1
R378H5	C833-5CMS x RZM R178 (C78/3)	11103	38.86	14.21	152	1.4	91.5	185	4.9
R378H3	C562HO x RZM R178 (C78/3)	8810	33.97	12.92	149	0.0	91.3	239	5.3
R378H62	2835-8H5 x RZM R178 (C78/3)	10608	39.04	13.56	156	0.4	92.1	216	5.0
R378H63	2835-10H5 x RZM R178 (C78/3)	9583	35.35	13.56	142	1.0	90.4	241	5.1
R378H64	2835-24H5 x RZM R178 (C78/3)	11010	39.84	13.73	154	0.4	92.3	211	4.9
R378H66	2836-13H5 x RZM R178 (C78/3)	9842	36.30	13.49	147	0.9	90.6	218	5.1
R378H67	0837-6H5 x RZM R178 (C78/3)	11456	40.33	14.15	155	0.4	91.7	194	4.8
R378H59	2869-15H5 x RZM R178 (C78/3)	9744	34.68	14.04	152	0.5	92.7	200	4.9
R378H60	2840-9H5 x RZM R178 (C78/3)	12080	45.14	13.34	148	0.9	93.5	243	5.1
R378H74	02-FC1015HO x RZM R178 (C78/3)	8993	32.69	13.84	154	9.7	90.7	227	5.1
R378H73	02-FC124HO x RZM R178 (C78/3)	9858	36.30	13.61	156	0.9	90.4	203	4.9
R378H42	C842HO x RZM R178 (C78/3)	9663	37.08	13.00	145	2.5	91.4	250	5.3
R378H92	C790H5 x RZM R178 (C78/3)	11219	40.31	13.00	154	0.0	81.9	189	4.8
Topcrosses with popn-931									
3931H5	C833-5CMS x RZM 2931	10692	39.30	13.60	159	1.2	92.4	184	4.9
3931H74	02-FC1015HO x RZM 2931	9011	31.68	14.12	152	25.4	92.4	201	4.9
3931H73	02-FC124 x RZM 2931	8749	32.55	13.46	154	1.7	90.2	191	4.8
3931H59	2869-15H5 x RZM 2931	10558	39.22	13.47	153	1.3	91.7	261	5.4
3931H62	2835-8H5 x RZM 2931	8812	32.95	13.38	159	0.5	89.7	205	4.9
3931H63	2835-10H5 x RZM 2931	10557	38.14	13.78	157	0.9	92.3	213	5.3

A131

TEST B104. EVALUATION OF TOPCROSS HYBRIDS, IMPERIAL VALLEY, CA, 2003-2004

(cont.)

Variety	Description	Acre Yield			Beets/		Clean	NO3-N	
		Sugar Lbs	Beets Tons	Sucrose %	100' No.	Bolters %	Beets %	ppm	score
Topcrosses with popn-931 (cont.)									
3931H64	2835-24H5 x RZM 2931	10217	38.44	13.28	159	1.2	92.0	212	5.0
3931H66	2836-13H5 x RZM 2931	9374	34.32	13.66	158	0.9	90.6	182	4.9
Mean		10290.1	37.62	13.62	154.0	2.3	91.2	217.3	5.0
LSD (.05)		1576.5	5.23	0.97	12.9	2.9	5.5	51.8	0.5
C.V. (%)		15.6	14.11	7.23	8.5	128.5	6.1	24.2	9.8
F value		4.0**	4.12**	1.31NS	1.3NS	25.8**	1.3NS	2.0NS	1.3NS

NOTES: 2931 = S^f,MM,A:aa population-931. H5 means that the female component was C833-5CMS.
        HO means near-CMS equivalent.

        Grown in soil without rhizomania or severe cyst nematode.

A132

TEST B204. EVALUATION OF HYBRIDS WITH S$_1$ PROGENY LINE POLLINATORS, IMPERIAL VALLEY, CA, 2003-2004

48 entries x 8 reps., RCB(E)
1-row plots, 18 ft. long

Planted: September 15, 2003
Harvested: May 20, 2004

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/ 100' No.	Bolters %	Clean Beets %	NO3-N ppm	score
Checks									
HH142	9-12-03	12449	44.79	13.90	173	1.2	93.4	209	5.1
Beta 4430R	8-21-03	12849	42.70	15.04	163	0.5	90.2	169	4.8
Phoenix	9-12-03	12361	44.50	13.90	163	0.0	92.8	206	4.9
Beta 4001R	8-25-03	14693	50.16	14.68	161	0.4	91.9	195	4.9
Populations hybrids									
3931H50	C790-15CMS x RZM 2931	10622	36.34	14.63	158	5.9	91.7	139	4.5
3941H50	x RZM 2941	10601	37.55	14.08	161	1.2	92.6	128	4.1
Z325H50	x RZM Z225,(Cz25)	10574	36.97	14.32	164	3.3	91.1	149	4.4
CR311H50	x RZM CR211	10506	37.27	14.09	165	6.4	91.9	206	5.0
3942H50	x RZM 2942	11877	40.50	14.63	163	0.0	92.7	143	4.4
3943H50	x 2943(C)	10441	36.20	14.44	164	1.7	91.6	135	4.3
N325H50	x RZM N224(C)(g)	10796	36.63	14.74	154	4.1	91.3	145	4.1
P207/8H50(Iso)	x RZM-PMR-NR P007/8,(CP07)	12736	43.97	14.45	170	1.2	92.8	126	4.1
Retests and new increases									
3927-4H50	C790-15CMS x RZM 9927-4,C927-4	11596	37.72	15.37	172	2.5	90.1	131	4.4
2930-19H50	x RZM 1930-19,C930-19	12531	42.75	14.68	163	0.0	91.2	149	4.4
Z325-9H50	x Z825-9,Cz25-9	12037	40.29	14.93	163	0.0	93.7	172	4.6
1929-62H50	x RZM 9929-62,C929-62	11968	42.34	14.11	166	0.0	91.2	189	4.8
2930-35H50	x RZM 1930-35,C930-35	11878	40.75	14.55	164	0.0	93.6	175	4.6
1929-4H50	x RZM 9929-4	13074	42.18	15.48	159	1.4	92.7	150	4.5
2929-45H50	x 9929-45	12478	42.64	14.65	168	3.8	93.4	142	4.4
1931-201H50	x 9931-201	13488	49.02	13.75	158	0.0	93.8	176	4.8

Variety	Description	Acre Yield Sugar Lbs	Beets Tons	Sucrose %	Beets/100' No.	Bolters %	Clean Beets %	NO3-N ppm	score
Retests and new increases (cont.)									
3931-56H50	C790-15CMS x RZM 1931-56	12372	42.69	14.49	166	0.8	92.2	112	3.9
Z331-14H50	x RZM Z131-14	11115	36.19	15.36	159	0.0	90.9	149	4.4
N330-5H50	x RZM N230-5-#(C)	10535	37.44	14.10	158	14.0	91.5	175	4.6
Angelina	3-10-03	12887	42.22	15.27	158	0.0	90.7	158	4.5
Selected S_1 lines									
3931-120H50	C790-15CMS x 1931-120	10814	37.57	14.47	167	0.4	90.4	137	4.3
3941-107H50	x 1941-107	10910	35.35	15.43	168	0.4	91.2	118	4.0
3941-112H50	x 1941-112	12180	39.57	15.38	162	0.8	90.9	104	4.1
3933-107H50	x 1933-107	10703	37.65	14.23	157	0.8	91.3	95	3.8
3933-113H50	x 1933-113	13722	46.93	14.62	155	0.0	91.8	169	4.6
3933-118H50	x 1933-118	13532	44.38	15.29	155	0.4	92.0	105	3.9
Z325-105H50	x Z125-105	10227	35.05	14.61	163	0.0	90.0	215	5.0
Z325-109H50	x Z125-109	10618	34.83	15.22	163	5.0	92.4	145	4.5
CR311-6H50	x CR111-6	10702	39.74	13.55	159	1.6	91.7	248	5.3
CR311-41H50	x CR111-41	11938	42.88	13.97	160	1.3	92.6	191	4.9
CR311-88H50	x CR111-88	13196	47.31	13.96	158	0.9	92.5	234	5.1
CR310-14-2H50	x RZM CR110-14-2	11756	41.64	14.13	160	0.0	91.2	197	4.9
03-FC1030-15H50	x 01-FC1030-15	10498	36.39	14.39	159	22.5	91.0	112	4.3
03-FC1030-16H50	x 01-FC1030-16	10939	39.16	13.96	163	2.2	92.3	150	4.4
Hybrids with C833-5CMS									
1927-4H5	C833-5CMS x RZM 9927-4,C927-4	12476	41.93	14.94	155	0.0	90.3	131	4.3
3931-56H5	x RZM 1931-56	12808	45.39	14.15	167	0.5	93.6	95	3.9
Z331-14H5	x RZM Z131-14	9872	31.41	15.70	158	0.0	91.6	160	4.5
2930-19H5	x RZM 1930-19,C930-19	11799	41.33	14.30	161	0.0	93.6	124	3.9

TEST B204. EVALUATION OF HYBRIDS WITH S₁ PROGENY LINE POLLINATORS, IMPERIAL VALLEY, CA, 2003-2004

(cont.)

Variety	Description	Acre Yield		Sucrose	Beets/100'	Bolters	Clean Beets	NO3-N	score
		Sugar Lbs	Beets Tons	%	No.	%	%	ppm	
Hybrids with C833-5CMS (cont.)									
2929-45H5	C833-5CMS x 9929-45	10948	38.28	14.32	142	1.7	92.5	117	4.0
1929-4H5	x RZM 9929-4	12716	40.61	15.68	166	1.6	93.7	144	4.4
P318-6H5	x P118-6, (CP08)	12271	40.73	15.04	165	0.4	92.5	104	4.0
P207/8H5	x P007/8, (CP07)	13166	44.80	14.67	153	0.0	93.4	119	4.1
N324H5	x RZM N224(g)	10886	37.07	14.69	165	0.0	92.6	90	3.6
Roberta	3-25-03	13007	49.30	13.28	159	0.4	93.5	129	4.3
Mean		11836.4	40.69	14.58	161.5	1.9	92.0	151.2	4.4
LSD (.05)		1476.0	4.93	0.72	15.0	2.8	2.5	43.0	0.5
C.V. (%)		12.7	12.31	5.04	9.4	150.0	2.8	28.9	12.2
F value		4.5**	5.53**	4.81**	1.0NS	15.6**	1.4*	6.0**	4.2**

NOTES: Grown in Field J that is not known to have significant rhizomania or sugarbeet cyst nematode (SBCN).
Fields J & K are adjacent and differ primarily in farming and cultural history. Field K has been
intentionally inoculated with rhizomania and SBCN, but rhizomania appears to be fairly light, SBCN fairly
heavy. No significant disease or pest problems were noted in Field J. H50 = C79-15CMS as seed bearing
parent. C790-15CMS = C790-68CMS x C790-15. H5 = C833-5CMS as seed bearing parent. S₁ progenies were
produced at Salinas and evaluated per se in tests at Salinas and Imperial Valley. Selected S₁'s were then
crossed to CMS's to produce experimental hybrids currently being tested in Imperial Valley and Salinas. S₁'s
were produced from MM,Sᶠ,A:aa populations undergoing population improvement.

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TEST B304. EVALUATION OF HYBRIDS WITH SELF-STERILE ($S^s s^s$) POLLINATORS, IMPERIAL VALLEY, 2003-2004

48 entries x 8 reps., RCB(E)
1-row plots, 18 ft. long

Planted: September 15, 2003
Harvested: May 21, 2004

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	Bolters %	Clean Beets %	NO3-N ppm	score
Checks									
Beta 4001R	8-25-03	14469	45.30	15.99	167	1.4	89.9	113	4.0
Phoenix	9-12-03	12183	42.17	14.43	158	0.0	94.1	136	4.3
Beta 4430R	8-21-03	13540	44.26	15.31	165	0.5	90.3	132	4.4
HH142	9-12-03	12029	40.98	14.69	165	2.2	93.1	117	4.1
Line hybrids									
R378H50	C790-15CMS x RZM R178, (C78/3)	11154	36.50	15.33	156	5.3	90.3	96	3.4
Y391H50	x RZM-ER-% Y191	11450	36.69	15.61	165	1.8	90.1	65	3.1
Y392H50	x RZM Y292	11565	37.91	15.30	165	3.8	92.4	97	3.9
Y375H50	x RZM Y275	11274	35.54	15.83	168	1.3	90.6	83	3.4
R321H50	x RZM R221, (C26,C27)	11133	37.52	14.80	168	3.9	91.8	102	3.9
P207/8H50(Sp)	x P007/8, (CP07)	10954	36.95	14.84	164	2.5	90.8	86	3.6
Retests and new increases									
P318-6H50(Sp)	C790-15CMS x P118-6, (CP08)	10642	34.84	15.22	174	0.8	88.0	97	3.9
P318-6H50(Iso)	x PMR-RZM P118-6, (CP08)	12224	39.89	15.34	157	1.5	92.2	94	3.9
Y367-5H50	x RZM Y167-5	10922	37.01	14.69	173	0.4	92.9	96	4.0
R243-14H50	x R043-14	11830	39.80	14.86	152	8.7	90.9	134	4.1
Y267-21H50	x Y067-21	10275	32.43	15.84	160	1.4	89.2	90	3.6
R280-6H50	x R080-6	11263	36.43	15.46	163	5.7	91.5	106	3.9
2930-19H50	C790-15CMS x RZM 1930-19, (C930-19)	12267	41.79	14.66	170	0.0	89.4	96	3.8
R381-22H50	x RZM R181-22, (C81-22)	10096	32.23	15.68	167	0.4	89.8	106	3.6
R378-6H50	x RZM R178-6	12385	41.08	15.10	161	0.0	91.2	94	3.6
R380-21H50	x RZM R180-21	12220	39.76	15.39	172	0.0	91.8	97	3.6
Y368-8H50	x RZM Y168-8	11586	38.24	15.16	167	1.2	91.0	113	4.0

TEST B304. EVALUATION OF HYBRIDS WITH SELF-STERILE (S^sS^s) POLLINATORS, IMPERIAL VALLEY, 2003-2004

(cont.)

Variety	Description	Acre Yield Sugar lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	Bolters %	Clean Beets %	NO3-N ppm	score
Selected FS lines									
Y390-40H50	C790-15CMS x Y190-40	11517	36.16	15.93	174	0.0	88.2	74	3.4
Y390-43H50	x Y190-43	10937	34.74	15.79	163	0.0	89.4	89	3.5
Y390-83H50	x Y190-83	12687	39.67	16.01	174	0.0	90.9	78	3.6
Y390-98H50	x Y190-98	12196	40.96	14.89	164	0.0	90.8	95	3.9
R376-89-10H50	x R176-89-10	11449	35.86	15.93	167	5.2	91.1	72	3.3
R376-89-4H50	x R176-89-4	11846	38.14	15.52	162	0.0	91.6	115	4.1
R376-89-5-4H50	x RZM R176-89-4-5, (C76-89-5-4)	11299	36.15	15.68	166	0.4	90.3	89	3.8
Y375-9H50	x Y175-9	12340	41.49	14.89	152	0.0	91.2	123	4.3
Y375-13H50	x Y175-13	11454	37.22	15.40	160	0.5	90.8	95	3.5
Y375-20H50	x Y175-20	10724	35.47	15.12	163	7.7	92.0	95	3.9
Roberta	3-25-03	12506	45.36	13.85	162	0.0	90.1	72	3.3
Hybrids with C833-5CMS									
R378H5	C833-5CMS x RZM R178,(C78/3)	13010	41.46	15.69	170	1.2	92.9	83	3.8
R381-22H5	x RZM R181-22,(C81-22)	11123	36.90	15.09	170	0.0	91.3	87	3.5
P318-6H5	x P118-6, (CP08)	13902	43.12	16.08	154	0.0	92.6	66	3.0
P207/8H5	x P007/8, (CP07)	12184	40.71	14.99	165	0.4	92.2	82	3.6
Y367-5H5	x RZM Y167-5	11323	37.33	15.18	166	0.0	92.3	76	3.3
Y378-6H5	x RZM Y178-6	12403	40.81	15.22	161	0.0	88.7	91	3.9
Y380-21H5	x RZM R180-21	12418	37.70	16.46	154	0.0	91.8	60	3.1
Y368-8H5	x RZM Y168-8	12305	39.59	15.57	157	0.0	91.6	78	3.6
Checks for rzm and SBCN tests									
USDA MM breeding lines									
N312	PMR-RZM-NR N112	9859	33.27	14.75	164	11.7	90.8	54	2.9
N372	RZM-ER-NR N172	8188	30.16	13.56	152	26.1	90.2	134	4.1

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TEST B304. EVALUATION OF HYBRIDS WITH SELF-STERILE (S^{s}S^s) POLLINATORS, IMPERIAL VALLEY, 2003-2004

(cont.)

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/ 100' No.	Bolters %	Clean Beets %	NO3-N ppm	score
USDA experimental hybrids									
1927-4H50	C790-15CMS x RZM 9927-4, (C927-4)	12571	40.93	15.37	165	1.2	91.1	86	3.6
R378H93	N265-31HO x RZM R178, (C78/3)	10822	37.29	14.55	163	2.5	92.2	134	4.1
Commercial hybrids									
Hil-1	4-22-03	9680	33.10	14.63	170	14.9	89.3	116	4.3
2VK0305	9-12-03	11378	40.82	13.93	160	0.0	89.7	143	4.5
2AP0852	9-12-03	12187	38.51	15.81	170	0.4	91.3	200	4.9
US H11	1999 production	8882	32.99	13.44	175	0.4	91.7	109	3.9
Mean		11596.9	38.19	15.18	164.1	2.4	91.0	98.8	3.8
LSD (.05)		1378.1	4.27	0.69	17.0	2.8	3.3	41.5	0.7
C.V.(%)		12.1	11.35	4.58	10.5	116.8	3.7	42.6	18.9
F value		5.5**	5.03**	6.94**	1.0NS	22.6**	1.2NS	3.0**	2.6**

NOTES: See notes for test B204. Prefix of "C" usually means lines has been officially released from Salinas, CA. Y = virus yellows et al. program. R = rhizomania et al. program. N = thought to have SBCN resistance either from *B.procumbens* or from *B.maritima*. P = is from powdery mildew et al. program including VY, rzm, SBCN, etc. Full-sib progenies were produced at Salinas from MM,S^{s}S^s lines undergoing population improvement. FS's were tested at Brawley and Salinas per se for NB, VY, rzm, PM, SBCN, etc. Selected FS's were increased and crossed to CMS's to produce experimental hybrids being evaluated here at Salinas. Some entries were included as Nondiseased checks for SBCN/rzm tests in Field K, particularly B504. N312 & N372 may have SBCN resistance from *Bvm*. N324 & N265-31HO have resistance from *B.procumbens*.

A138

TEST B404. HYBRIDS WITH COMBINED RESISTANCE TO RHIZOMANIA & SBCN, IMPERIAL VALLEY, 2003-2004

Planted: September 15, 2003
Harvested: May 24, 2004

24 entries x 8 reps., RCB(E)
1-row plots, 18 ft. long

Variety	Description			Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	Clean Beets %	NO3-N ppm	score	Appearance Mean
Checks											
Beta 4430R	Rz	8-21-03		4731	14.36	16.56	165	93.6	126	4.1	2.9
Phoenix	Rz	9-12-03		3843	12.68	15.15	172	94.0	228	4.9	3.6
Topcrosses with C78/2 & Y191											
R378H50	Rz	C790-5CMS	x RZM R178, (C78/3)	2573	8.26	15.81	167	90.8	188	4.9	3.1
R378H5	Rz	C833-5CMS	x RZM R178	3034	9.11	16.67	151	91.1	114	4.0	2.7
R378H68	Rz,R22	2848-1H5	x RZM R178	3642	11.31	16.07	156	89.9	137	4.4	2.6
R378H80	Rz,R22	2810-17H5	x RZM R178	3875	12.82	15.19	165	93.6	187	5.0	2.3
R378H81	Rz,R22	2810-19H5	x RZM R178	3762	11.70	16.07	153	92.9	121	4.1	2.5
R378H97	Rz,Bp	N267HOg	x RZM R178	3417	11.29	15.18	161	89.6	194	4.9	2.3
R378H99	Rz,Bp	N265(C)HOg	x RZM R178	2641	9.04	14.60	152	89.3	188	4.8	3.1
R378H93	Rz,Bp	N265-31HOg	x RZM R178	3546	11.57	15.32	155	88.8	172	4.6	2.4
R378H94	Rz,Bp	N265-9HOg	x RZM R178	4004	13.81	14.49	152	87.6	189	4.6	2.5
Y391H50	Rz,R22	C790-15CMS	x RZM -ER-% Y191	4361	13.09	16.69	163	91.9	132	4.3	2.2
Nematode resistant pollinators											
N325H50	Rz,Bp	C790-15CMS	x RZM N224(C) (g)	3081	10.00	15.42	165	88.0	164	4.6	3.1
N330-5H50	Rz	C790-15CMS	x RZM N230-5-#(C) (g)	909	3.27	13.86	165	86.2	188	4.8	4.0
3927-4H50	Rz,R22	C790-15CMS	x RZM 2927-4, (C927-4)	7180	22.21	16.17	165	94.5	169	4.9	1.3
1927-4H5	Rz,R22	C833-5CMS	x RZM 9927-4, (C927-4)	6684	21.12	15.84	149	94.7	142	4.4	1.6

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TEST B404. HYBRIDS WITH COMBINED RESISTANCE TO RHIZOMANIA & SBCN, IMPERIAL VALLEY, 2003-2004

(cont.)

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	Clean Beets %	NO3-N ppm	Appearance score	Appearance Mean
Nematode resistant pollinators (cont.)									
P207/8H50(Sp) Rz,WB242	C790-15CMS x P007/8, (CP07)	4596	14.25	16.17	162	93.8	145	4.5	2.0
P207/8H5 Rz,WB242	C833-5CMS x P007/8, (CP07)	4958	15.12	16.31	159	95.2	112	4.0	2.1
P318-6H50(Sp) Rz,R22,WB242	C790-15CMS x P118-6, (CP08)	4648	14.55	16.00	155	90.9	121	4.3	1.9
P318-6H5 Rz,R22,WB242	C833-5CMS x P118-6, (CP08)	5621	17.00	16.53	166	93.5	86	3.5	1.8
Y375H50 Rz,R22	C790-15CMS x RZM Y275	6099	19.27	15.94	165	93.2	184	4.8	1.7
Y375-9H50 Rz,R22	C790-15CMS x Y175-9	5314	16.29	16.32	165	90.9	158	4.4	2.4
Y367-5H5 Rz,R22	2833-5CMS x RZM Y167-5	4535	14.13	16.07	153	94.8	136	4.5	1.9
Susceptible check									
USH11	susceptible check	1995	6.75	14.75	165	87.8	122	4.3	3.5
Mean		4127.0	13.04	15.72	160.3	91.5	154.4	4.5	2.5
LSD (.05)		949.4	3.03	0.68	11.9	2.7	44.6	0.5	0.5
C.V. (%)		23.4	23.57	4.35	7.5	3.0	29.4	12.2	20.0
F value		18.1**	16.01**	9.80**	2.2NS	7.5**	4.7**	3.6**	14.9**

NOTES: Appearance score: (= beauty score) rating of canopy prior to harvest. Mean scores were from ratings made 5/19/04 and 5/21/04, where 1 = best and 5 = worst. 1 = estimate of how canopy (size, color, bigor, wilting, Chlorosis, necrosis, survival, etc.) would look in the absence of disease (rhizomania and cyst nematode). 5 = plants stunted, dead, dying and in very poor general health. 3 = approximately how lines with only Rz factor would rate.

Tests B404-B704 were grown in Field K. Tests B104-B304 in Field J. Field J is relatively healthy. Field K has been inoculated with SBCN & BNYVV (pathotype-A) and sugarbeet is grown on a 2-year rotation. Results suggest that rhizomania is moderate and SBCN severe. C790-15CMS is *rzrz*. C833-5CMS is *RzRz*. All B.p. sources segregate.

A140

TEST B504. PERFORMANCE OF COMMERCIAL AND EXPERIMENTAL HYBRIDS UNDER RHIZOMANIA AND SBCN CONDITIONS, IMPERIAL VALLEY, 2003-2004

24 entries x 8 reps., RCB(E)
2-row plots, 18 ft. long

Planted: September 15, 2003
Harvested: May 25, 2004

Variety	Resistance Rz NR1 NR2	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	Clean Beets %	Bolting %	NO3-N ppm	NO3-N score	Appear Mean
Checks											
Beta 4430R	√	3-21-03	4864	15.19	16.15	165	94.2	0.0	133	4.3	3.0
Phoenix	√	9-12-03	4504	14.87	15.23	161	95.2	0.0	180	4.5	3.8
Roberta		3-25-03	5388	16.39	16.62	159	94.3	0.0	143	4.3	3.5
US H11		1999 production	2297	8.22	13.87	159	90.9	0.0	112	3.9	3.7
Syngenta hybrids											
Hil-1	√ √	4-22-03	8011	24.01	16.69	155	91.8	0.0	122	4.1	2.5
Hil-2	√ √	4-22-03	8773	26.79	16.37	157	92.6	0.0	151	4.6	2.4
Hil-3	√ √	4-22-03	7665	25.90	14.77	160	92.2	0.0	141	4.4	2.7
USDA MM breeding lines											
N312	√ √ √	PMR-RZM-NR N112	6129	20.62	14.85	152	93.7	0.7	116	3.9	1.3
N372	√ √ √	RZM-ER-NR N172	5758	19.91	14.52	146	93.5	20.8	179	4.7	2.3
Betaseed hybrids											
2VK0305	√ √	9-12-03	10410	31.48	16.58	161	94.3	0.2	146	4.4	2.1
OVK6280	√ √	9-12-03	6732	21.85	15.53	158	89.6	0.0	118	4.0	2.2
2AP0852	√ √	9-12-03	10844	31.26	17.35	164	96.1	0.0	162	4.5	1.3
2EN5066	√ √	9-12-03	9005	28.79	15.66	167	94.0	0.0	104	4.0	1.8
USDA Experimental hybrids											
1927-4H50	√ √	C790-15CMS x RZM 9927-4, (C927-4)	6862	22.05	15.57	151	94.5	0.0	123	4.1	1.8
3931-56H50	√ √	C790-15CMS x 1931-56	2470	8.14	15.24	156	91.8	0.0	87	3.8	4.1
P207/8H50	√	C790-15CMSxP007/8, (CP07)	4737	14.80	15.96	156	94.3	0.0	116	4.0	2.0
N330-5H50	√	C790-15CMS x RZM N230-5-#(C)(g)	1350	4.88	13.81	155	88.2	0.0	115	3.9	4.1
N325H50	√	C790-15CMS x RZM N224(C)(g)	4813	15.24	15.82	158	90.0	0.0	95	3.8	2.4

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TEST B504. PERFORMANCE OF COMERCIAL AND EXPERIMENTAL HYBRIDS UNDER RHIZOMANIA AND SBCN CONDITIONS, IMPERIAL VALLEY, 2003-2004

(cont.)

Variety	Resistance Rz	NR1	NR2	Description	Acre Yield Sugar Lbs	Beets Tons	Sucrose %	Beets/100' No.	Clean Beets %	Bolting %	NO3-N ppm	score	Appear Mean
USDA Experimental hybrids (cont.)													
R378H93	√	√		N265-31HO x RZM R178, (C78/3)	4275	13.96	15.28	152	91.4	0.0	127	4.1	2.5
N224H98,94	√	√		{N165-9H50(g) x RZM-NR N124(g) {N165-9HO(g) x N124-#(C)(g)	4901	15.79	15.54	147	88.4	0.0	95	3.8	2.7
Y375H50	√			C790-15CMS x RZM Y275	6267	19.26	16.24	157	93.0	0.0	104	4.0	1.9
Y367-5H50			√	C790-15CMS x RZM Y167-5	4458	13.50	16.54	159	94.4	0.0	77	3.5	2.5
Y367			√	RZM-ER-% Y167,(C672)	4754	14.02	16.95	154	94.0	0.0	62	3.1	2.4
R381-22H50	√			C790-15CMS x RZM R181-22,(C81-22)	2556	7.59	16.84	154	91.9	0.0	95	3.6	3.4
Mean					5742.6	18.11	15.75	156.7	92.7	0.9	120.8	4.1	2.6
LSD (.05)					773.9	2.30	0.77	8.3	1.5	1.6	40.8	0.6	0.5
C.V. (%)					13.7	12.87	4.95	5.4	1.6	178.6	34.3	14.4	17.7
F value					79.4**	80.79**	11.69**	3.0*	15.4**	55.3**	4.1**	3.4**	24.9**

Rz = Holly (Rz1) or other sources of resistance to rhizomania
NR1 = Beta procumbens source of nematode resistance.
NR2 = Possibly other sources of nematode resistance or tolerance.

See tests B104-B304 for relative nondiseased conditions.

NOTES: Appearance score: (= beauty score) rating of canopy prior to harvest. Mean scores were from ratings made 5/19/04 and 5/21/04, where 1 = best and 5 = worst. 1 = estimate of how canopy (size, color, vigor, wilting, chlorosis, necrosis, survival, etc.) would look in the absence of disease (rhizomania and cyst nematode). 5 = plants stunted, dead, dying and in very poor general health. 3 = approximately how lines with only Rz factor would rate.

(cont.)

SUGARBEET CYST NEMATODE SOIL COUNTS

Variety	Resistance			Description	Cysts Full/Viable		No. Eggs & Larvae	
	Rz	NR1	NR2		3/04	5/23	3/04	5/23
Beta 4430R	✓			3-21-03	86.8	117.0	10590.0	12417.0
Phoenix	✓			9-12-03	94.1	171.5	12716.3	16215.8
Hil-2	✓	✓		4-22-03	24.5	27.4	1413.8	2345.6
N312	✓		✓	PMR-RZM-NR N112	59.9	103.5	5536.9	6628.9
N372	✓		✓	RZM-ER-NR N172	61.9	73.0	6485.6	5511.0
2VK0305	✓	✓		9-12-03	21.8	36.8	1869.4	3216.4
2AP0852	✓		✓	9-12-03	28.8	45.3	1710.0	2651.6
1927-4H50	✓		✓	C790-15CMS x RZM 9927-4, (C927-4)		171.8		15131.3
Y367	✓		✓	RZM-ER-% Y167, (C672)	86.5	233.4	12210.0	22714.5
Mean					58.0	107.0	6566.5	9648.0
LSD (.05)					18.3	48.7	2650.6	3735.0
C.V. (%)					31.4	45.5	40.2	38.7
F value					21.4**	15.4**	26.1**	30.3**

March 2004 readings (8 varieties x 8 reps); May 2004 readings (9 varieties x 8 reps)

NOTES: Counts for 100 grams soil. Soil cores (samples) taken 12 inches deep, 8 cores/plot composited, 3-4 inches
from plants on inside shoulder of beds, 2-row plots. On 3/04/04, 8 entries were sampled. On 5/23/04, at harvest,
9 entries were sampled. Initial samples and counts were not made.

(cont.)

GREENHOUSE CYST COUNTS

Variety	Resistance Rz	NR1	NR2	Description	No. Days	No. Plants	No. Cysts range	mean	No. Plants ≤ 12 cysts
Beta 4430R	√			3-21-03	48	1	21		
					125	1	83		
					49	1	71		
					126	1	22		
Hil-2	√	√		4-22-03	48	1	0		
					125	1	2		
					49	1	0		
					126	1	1		
N312	√		√	PMR-RZM-NR N112	48	12	4- 48	27	5
					125	10	1-194	43	3
N372	√		√	RZM-ER-NR N172	49	10	1- 76	21	5
					126	10	9-246	55	4
2AP0852	√		√	9-12-03	48	1	5		
					125	1	5		
					49	1	9		
					126	1	1		
Angelina					48	1	16		
					125	1	75		
					49	1	107		
					126	1	42		
OVK6280					48	1	5		
					125	1	6		
					49	1	1		
					126	1	2		

TEST B504. PERFORMANCE OF COMMERCIAL AND EXPERIMENTAL HYBRIDS UNDER RHIZOMANIA AND SBCN CONDITIONS,
IMPERIAL VALLEY, 2003-2004

(cont.)

NOTES: N312 & N372 are MM,S^f,A:aa lines. During their development for resistance to rhizomania, powdery mildew, and other diseases, it was observed in field tests, mother root selections and progeny tests at Salinas and Brawley that some plants did not have infection with sugarbeet cyst nematode (SBCN). Non-infested plants were selected and increased. These field observations were not controlled, however, and subject to escapes, cycling of cyst development, field variability, etc. Only in 2004 have controlled greenhouse tests been run at Salinas on individual plants from these lines. For the conetainer tests in the greenhouse, soil from Spence field, known to have both SBCN and rhizomania, was used. After being germinated in sand, individual seedlings were transplanted to conetainers. Total cysts were counted within each conetainer. N312 (line N12) has WB242 & 97 (B.vulgaris subsp. maritima) germplasm. N372 (line N72) has Bvm germplasm accessed in 1993 from a European source.

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TEST B604. PERFORMANCE OF LINES WITH RESISTANCE TO RHIZOMANIA AND SBCN, IMPERIAL VALLEY, 2003-2004

Planted: September 15, 2003
Harvested: May 26, 2004

48 entries x 6 reps., RCB
1-row plots, 18 ft. long

Variety	Resistance	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	Clean Beets %	Bolting %	NO3-N ppm	Appear Mean	SBCN Reaction
Checks											
Phoenix	Rz	9-12-03	3902	13.23	14.76	159	94.5	0.0	191	3.8	
Beta 4430R	Rz	8-21-03	4427	13.89	16.07	168	92.7	0.0	119	3.2	1/32
Angelina	Rz1,Rz2		2747	9.59	14.66	163	89.2	0.0	145	3.5	0/38
Roberta	--		4422	13.25	16.83	170	92.7	0.0	155	3.3	
US H11	--	1999 production	1947	6.90	14.10	155	86.2	0.0	143	3.5	
1927-4H5	Rz,R22	C833-5CMS x RZM 9929-4, (C927-4)	7235	22.85	15.70	158	91.3	0.0	124	1.4	
R378H93	Rz,Bp	N265-31HO x RZM R178, (C78/3)	4561	15.53	14.71	152	87.9	0.0	167	2.1	
P318-6H5	Rz,WB242	2833-5HOxP118-6, (CP08)	6543	19.52	16.78	153	91.7	0.0	66	2.1	6/10
Multigerm breeding lines											
R378(Iso)	Rz	RZM-ER-% R178, (C78/3)	2792	8.67	15.95	165	88.4	0.0	119	3.3	
Y390	Rz	Inc. Y190-#(C),C2,Syn1	2745	8.81	15.30	157	89.0	0.0	129	3.6	
Y391	Rz,R22	RZM-ER-% Y191,C2,Syn1	3258	10.28	15.94	156	94.1	0.7	101	2.9	
Y392	Rz,R22	RZM Y292,C1,Syn2	4158	14.00	14.68	157	90.1	0.0	162	2.2	
Y393	Rz,R22	RZM FS-#(C),C1,Syn1	3624	11.93	15.31	150	93.1	1.2	115	2.9	
Y375	Rz,R22	RZM Y275	6295	20.40	15.40	168	93.4	1.0	140	1.9	9/21
Y321	Rz,Bvm	RZM R221, (C26,C27)	4813	17.35	13.94	154	94.0	4.2	183	2.3	
Y367	Rz,R22	RZM-ER-% Y167,(C67/2)	4113	12.43	16.63	161	93.0	0.0	63	2.3	3/25
Y371	Rz,R22	RZM-ER-% Y171	4431	15.47	14.41	159	92.9	0.0	129	2.6	1/10
03-C37	--	Inc. U86-C37	1416	5.12	13.71	163	86.8	0.0	103	4.5	
P328	Rz,WB242	PMR-RZM P228, (CP04)	6419	23.27	13.78	166	89.0	0.6	153	1.4	0/15
P327	Rz,WB97	PMR-RZM P227, (CP03)	1313	5.00	13.10	157	80.4	0.0	59	4.1	
R336	R22	RZM-ER-% R136, (C79-8)	3645	12.91	14.04	160	93.3	2.6	193	2.7	4/35
R340	R22	RZM-ER-% R140	3366	11.76	14.31	171	90.8	2.2	130	3.2	1/5
R324/5	WB41/42	Inc. R824, (C79-2,-3)	1502	5.21	14.61	158	82.5	0.0	82	3.9	
R337	WB151	Inc. R637,(C79-9,WB151)	1162	4.49	13.01	150	87.9	0.0	143	4.0	

TEST B604. PERFORMANCE OF LINES WITH RESISTANCE TO RHIZOMANIA AND SBCN, IMPERIAL VALLEY, 2003-2004

(cont.)

Variety	Resistance	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	Clean Beets %	Bolting %	NO3-N ppm	Appear Mean	SBCN Reaction
Multigerm breeding lines (cont.)											
R381-22	Rz	RZM R181-22,(C81-22)	2500	7.79	16.20	151	91.9	0.0	100	3.6	
R343	R22	RZM-ER-% R143	4045	13.74	14.56	171	92.2	3.8	105	2.2	2/10
R243-14	R22	Inc. R043-14	4430	14.77	14.75	153	90.1	0.0	94	2.4	0/5
3927-4	R22,Rz	RZM 2927-4(A,aa),(C927-4)	4833	16.06	15.14	166	93.7	0.0	118	2.3	20/30
N372	Rz,Bvm	RZM-ER-NR N172	6018	21.43	13.97	167	92.5	9.6	198	1.8	16/30
N312	Rz,WB242	PMR-RZM-NR N112	5269	17.70	14.93	156	87.7	2.7	110	1.7	16/41
P329	Rz,WB97	PMR-RZM P229,(CP05)	1995	6.89	14.44	154	86.7	0.0	127	4.1	
P330	Rz,WB242	PMR-RZM P230,(CP06)	2979	10.03	14.57	168	91.2	0.0	129	3.7	0/9
R378(Iso)	Rz	RZM-ER-% R178, (C78/3)	2427	7.77	15.60	167	88.3	0.0	123	3.7	
P307/8	Rz,WB97/242	PMR-RZM P207/8,(CP07)	6214	20.12	15.56	151	92.8	0.0	119	2.0	2/18
P207/8(Iso)	Rz,WB97/242	RZM-PMR-NR P007/8,(CP07)	5269	17.28	15.25	163	93.4	0.0	95	1.9	
N172	Rz,Bvm	NR-RZM N972 (A,aa)	5924	22.74	13.17	156	93.3	5.8	302	2.3	
Multigerm breeding lines											
N324	Rz,Bp	RZM N224(g)	3149	11.15	14.03	155	88.1	0.0	82	3.3	20/25
N325	Rz,Bp	RZM N224(C)(g)	3336	10.59	15.83	159	90.2	0.6	64	3.3	9/10
P318-6(Iso)	Rz,WB97/242	PMR-RZM P118-6,(CP08)	6346	20.43	15.50	152	90.9	0.0	70	1.3	3/25
P318-6(Sp)	Rz,WB97/242	Inc. P118-6,(CP08)	5722	18.21	15.66	162	92.8	0.0	68	1.7	
2927-4	R22,Rz	RZM 1927-4,(C927-4)	5991	19.33	15.50	159	92.7	0.0	113	1.6	20/30
Y267-21	R22,Rz	Inc. Y067-21	5842	17.88	16.35	163	94.6	0.0	80	1.9	0/5
Y271-14	R22,Rz	Inc. Y071-14	2675	8.17	16.44	175	90.1	0.0	52	2.8	0/5
Y367-5	R22,Rz	RZM Y167-5	3878	13.39	14.43	155	92.9	0.0	89	2.9	0/5
Y375-9	Rz,R22	Inc. Y175-9	4939	17.05	14.47	156	94.7	0.0	147	2.7	1/5
Y375-13	Rz,R22	Inc. Y175-13	4691	16.12	14.52	158	91.9	0.0	107	1.9	1/5
Y375-20	Rz,R22	Inc. Y175-20	6058	19.52	15.51	157	93.7	1.4	102	1.5	2/5
3931	Rz	RZM 2931,1931aa x A	1956	6.82	14.34	147	87.0	0.0	116	4.3	

TEST B604.   PERFORMANCE OF LINES WITH RESISTANCE TO RHIZOMANIA AND SBCN, IMPERIAL VALLEY, 2003-2004

(cont.)

| Variety | Resistance | Description | Acre Yield | | Sucrose | Beets/ 100' | Clean Beets | Bolting | NO3-N | Appear | SBCN |
| | | | Sugar | Beets | | | | | | | |
			Lbs	Tons	%	No.	%	%	ppm	Mean	Reaction
Mean			4110.9	13.68	14.97	159.3	90.8	1.0	121.3	2.7	
LSD (.05)			1307.9	4.14	1.15	18.0	5.1	2.5	56.3	0.6	
C.V. (%)			28.0	26.57	6.73	10.0	5.0	223.3	40.9	18.7	
F value			11.8**	12.76**	5.43**	1.0NS	2.9**	11.8**	4.9**	18.1**	

NOTES: MM lines undergoing improvement and development at Salinas. sources of resistance: Rz = Rz1. Rz2 = WB42. R22 = breeding line number for C50,C51 releases with Bvm background. Bp = nematode resistance from B.procumbens. WB242 = Bvm and possible source of resistance to PM and SBCN. WB97 = possible PMR source. WB41,42,151 with resistance to rzm accessed from Denmark. N172,N372,N112 & N312 have been intentionally selected for resistance to SBCN from Bvm. N324,N325 are MM,S^f,A:aa popns that segregate for Hs from B.procumbens.

APPEARANCE SCORE: Appearance score = canopy beauty score prior to harvest, where 1 = best and 5 = worst. 1 = estimate of how canopy (size, color, vigor, wilting, chlorosis, necrosis, survival, etc.) would look in the absence of disease (rhizomania & SBCN). 5 = plants stunted, dead, dying and in very poor general health. 3 = approximately how lines with only Rz1 factor would rate.

SBCN REACTION: Based upon greenhouse tests in 2004 at Salinas.

GREENHOUSE TESTS: N312 & N372 are MM,S^f,A:aa lines. During their development for resistance to rhizomania, powdery mildew, and other diseases, it was observed in field tests, mother root selections and progeny tests at Salinas and Brawley that some plants did not have infection with sugarbeet cyst nematode (SBCN). Non-infested plants were selected and increased. These field observations were not controlled, however, and subject to escapes, cycling of cyst development, field variability, etc. Only in 2004 have controlled greenhouse tests been run at Salinas on individual plants from these lines. For the container tests in the greenhouse, soil from Spence field, known to have both SBCN and rhizomania, was used. After being germinated in sand, individual seedlings were transplanted to containers. Total cysts were counted within each container. N312 (line N12) has WB242 & 97 (B.vulgaris subsp. maritima) germplasm. N372 (line N72) has Bvm germplasm accessed in 1993 from a European source.

Plants with < 12 cysts/total plants checked. Total cysts counted on plant and in soil of each container. Scores for checks in these greenhouse tests were: Beta 4430R 1/32, Angelina 0/38, Hil-2 39/42, OVK6280 25/27, and 2AP0852 27/35.

TEST B704. PROGENY PERFORMANCE UNDER RHIZOMANIA AND SBCN, IMPERIAL VALLEY, 2003-2004

Planted: September 15, 2003
Harvested: May 26, 2004

92 entries x 2 reps., RCB(E)
1-row plots, 18 ft. long

Variety	Resistance	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	Clean Beets %	Bolting %	NO3-N ppm	NO3-N score	Appear Mean
Checks											
Beta 4430R	Rz	8-21-03	4464	14.47	15.31	189	80.1	0.0	296	5.5	2.8
Phoenix	Rz	9-12-03	4392	15.05	14.41	175	82.9	0.0	354	6.0	3.3
US H11		1999 production	2588	9.48	13.51	161	89.0	0.0	309	5.5	3.5
Roberta		Susc. check	6340	18.35	17.31	175	94.7	0.0	276	5.5	3.8
1927-4H5	Rz,R22	C833-5HO x RZM 9927-4,(C927-4)	7205	24.78	14.81	139	94.6	0.0	239	5.5	2.0
Y393	Rz,R22	Inc. FS-#(C),C1,Syn1	4337	17.15	13.41	153	91.7	20.4	345	5.5	2.5
P318-6(Iso)	Rz,R22,WB242	PMR-RZM P118-6,(CP08)	4962	16.60	15.08	158	80.0	1.7	221	5.0	1.8
P207/8(Iso)	Rz,WB242	RZM-PMR-NR P007/8,(CP07)	5606	20.20	14.07	164	80.6	2.0	270	5.5	1.8
Nemtode resistant lines											
N312	Rz,WB242	PMR-RZM-NR N112	4212	14.91	13.97	164	88.6	.5.2	296	5.5	1.5
N372	Rz,Bvm	RZM-ER-NR N172	5200	20.61	12.22	167	86.4	26.1	518	6.5	2.0
N324	Rz,Bp	RZM N224(g)	3972	16.45	11.91	147	93.0	0.0	342	5.5	2.8
N325	Rz,Bp	RZM N224(C)(g)	3802	13.29	14.43	153	89.8	5.5	217	5.0	3.0
N369	Rz,Bp	RZM N269-#(C)(g)	3317	11.56	14.19	145	95.3	0.0	357	5.5	3.5
N365-31HO	Rz,Bp	RZM N265-31HO(g) x RZM N265-31(g)	4708	20.93	11.60	145	91.2	0.0	493	6.5	2.5
N365	Rz,Bp	RZM N265(g)	2789	15.62	9.23	167	81.8	0.0	720	7.0	2.8
N366	Rz,Bp	RZM N265(C)(g)	2281	11.17	10.80	175	79.6	0.0	558	6.5	3.3
FS progeny from Y75											
Y375	Rz,R22	RZM Y275	4464	15.83	14.14	158	87.8	5.2	271	5.5	1.8
Y375-301		RZM Y275 PX	5882	20.67	14.17	164	85.5	13.6	272	5.5	1.5
-302			5350	19.02	14.11	133	93.5	0.0	220	5.0	2.3
-303			6043	21.20	14.14	136	93.7	0.0	341	6.0	1.3

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(cont.)

Variety	Resistance	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/100' No.	Clean Beets %	Bolting %	NO3-N ppm	NO3-N score	Appear Mean
FS progeny from Y75 (cont.)											
Y375 -304	Rz,R22	RZM Y275 PX	5862	22.18	13.32	139	95.7	3.8	532	6.5	2.0
-305			8409	30.12	14.14	167	87.1	0.0	316	5.5	1.5
-306			4188	18.73	12.05	161	91.0	0.0	363	5.5	3.5
-307			7417	26.69	14.01	158	89.5	0.0	343	6.0	1.5
-308			6235	22.63	13.84	142	94.2	19.2	205	5.0	1.8
-309			4962	17.70	14.03	142	81.6	0.0	327	5.5	2.5
-310			6701	22.77	14.75	139	93.2	0.0	181	4.5	2.0
-311			8185	26.32	15.55	147	95.4	0.0	175	4.5	1.3
FS progenies from CP07											
P307/8	Rz,WB242	PMR-RZM P207/8(Iso),(CP07)	5033	17.60	14.33	142	95.7	0.0	213	5.0	1.5
P307 -301		PMR-RZM P207/8 PX	6147	22.13	14.39	150	94.9	0.0	203	4.5	2.3
-302			5953	25.37	11.74	150	88.5	0.0	313	6.0	1.3
-303			4486	16.79	13.61	145	78.4	0.0	209	5.0	1.3
-304			2418	8.33	14.59	158	92.6	1.6	205	5.0	2.5
-305			4743	15.35	15.55	156	88.0	0.0	105	3.5	1.5
-306			7349	25.01	14.69	153	92.3	0.0	165	4.5	1.5
-307			7238	26.71	13.54	158	96.8	0.0	275	5.5	1.5
-308			6442	22.83	14.12	139	96.5	0.0	285	5.5	1.0
-309			4864	18.61	13.56	150	94.8	0.0	257	5.5	2.3
-310			4105	14.57	14.11	145	79.8	0.0	247	5.0	2.8
-311			5307	17.73	15.32	139	88.0	0.0	193	4.5	1.8
-312			3487	12.43	13.91	153	87.1	0.0	216	5.0	2.5
-313			5624	20.35	14.05	147	82.0	0.0	317	5.5	2.8
-314			3518	12.46	13.92	153	92.0	0.0	187	5.0	3.5
-315			6674	24.37	13.67	145	96.8	0.0	262	5.5	2.3

TEST B704. PROGENY PERFORMANCE UNDER RHIZOMANIA AND SBCN, IMPERIAL VALLEY, 2003-2004

(cont.)

Variety	Resistance	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Acre Yield Sucrose %	Beets/ 100' No.	Clean Beets %	Bolting %	NO3-N ppm	score	Appear Mean
FS progenies from CP05											
P329	Rz,WB97	PMR-RZM P229,(CP05)	2927	10.97	13.03	153	92.3	2.0	369	6.0	3.8
P329-301		PMR-RZM P229PX	3442	12.31	13.87	170	86.3	0.0	231	5.0	3.8
-302			3559	14.32	12.51	156	83.3	0.0	312	5.5	3.3
-303			4547	16.61	13.79	125	88.9	0.0	402	6.0	3.5
-304			5721	20.82	13.43	175	84.8	0.0	318	5.5	2.8
-305			3470	10.97	15.76	150	83.2	0.0	128	4.0	3.0
-306			4887	16.45	14.94	139	83.6	0.0	312	5.5	2.8
P328	Rz,WB242	PMR-RZM P228,(CP04)	5324	19.99	13.29	147	93.9	11.3	255	5.0	1.5
FS progenies from CP06											
P330	Rz,WB242	PMR-RZM P230,(CP06)	4012	13.48	15.00	153	92.9	3.7	189	5.0	3.3
P330-301		PMR-RZM P230PX	5064	17.30	14.69	156	92.7	0.0	243	5.0	3.0
-302			4540	14.33	16.10	158	78.2	0.0	267	5.5	2.5
-303			3693	12.88	14.20	167	92.8	0.0	543	6.5	2.8
-304			3920	12.39	15.72	161	78.1	0.0	171	4.5	2.3
-305			3743	13.75	13.80	156	77.0	0.0	202	4.5	2.0
-306			4431	15.84	14.09	156	93.3	0.0	229	5.0	1.8
-307			4857	16.13	15.02	161	92.5	0.0	160	5.0	3.3
-308			4286	14.61	14.51	164	94.3	0.0	182	5.0	3.5
-309			3128	10.81	14.63	136	89.5	0.0	370	6.0	3.5
-310			3063	12.44	12.66	156	71.2	11.7	264	5.0	4.0
-311			2202	8.55	13.09	145	86.8	0.0	394	5.5	3.5
-312			2370	8.72	13.66	150	76.1	12.5	246	5.0	2.5
Sn progenies from C927-4											
3927-4	Rz,R22	RZM 2927-4(A,aa),(C927-4)	6408	23.69	13.44	153	84.7	3.1	276	5.5	2.3
3927-4-301		RZM 2927-4⊗	7800	28.15	13.85	136	96.1	0.0	240	5.5	2.3
-302			6447	21.49	15.00	128	97.2	0.0	123	4.0	1.5

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TEST B704. PROGENY PERFORMANCE UNDER RHIZOMANIA AND SBCN, IMPERIAL VALLEY, 2003-2004

(cont.)

Variety	Resistance	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Beets/ 100' No.	Clean Beets %	Bolting %	NO3-N ppm	NO3-N score	Appear Mean
Sₙ progenies from C927-4 (cont.)											
3927-4 -303		RZM 2927-4⊗	7018	24.49	14.38	145	95.4	0.0	153	4.5	2.3
-304			7490	27.99	13.45	156	94.6	0.0	228	5.0	2.5
-305			3941	13.61	15.03	156	72.2	0.0	137	4.5	3.0
-306			5285	21.32	12.41	150	83.3	0.0	284	5.5	2.5
2927-4 Rz,R22		RZM 1927-4(A,aa),(C927-4)	6181	22.09	13.98	167	90.7	0.0	245	5.5	1.5
3927-4 -307		RZM 2927-4⊗	3631	15.62	12.13	136	76.3	0.0	291	5.5	3.5
-308			9603	34.03	14.11	133	97.0	0.0	264	5.5	1.8
-309			3308	12.81	13.07	150	92.4	0.0	130	4.5	3.5
-310			5915	22.71	13.02	128	95.8	0.0	255	5.0	1.8
-311			5197	20.20	12.33	142	93.4	0.0	304	5.5	3.8
-312			4944	18.11	13.64	145	84.1	0.0	193	5.0	2.3
-313			7303	27.54	13.25	147	88.8	10.3	229	5.0	2.0
-314			4511	16.93	13.33	161	89.2	0.0	288	5.5	2.0
-315			8908	33.18	13.49	153	96.7	0.0	240	5.0	1.3
-316			3637	19.54	9.23	139	93.3	0.0	584	6.5	2.8
-317			6729	24.82	13.56	111	97.3	0.0	467	6.0	2.3
-318			6693	22.46	14.90	142	95.6	0.0	196	5.0	1.3
1927-4	Rz,R22	RZM 9927-4aa x A, (C927-4)	8507	31.25	13.60	97	94.2	0.0	273	5.5	1.3
S₁ progenies from popn-921											
3921 -301	Rz,Bvm	RZM 2921⊗	3186	12.98	12.44	145	77.5	0.0	303	5.0	3.3
-302			5005	18.61	13.45	156	90.5	0.0	280	5.5	2.3
-303			3515	12.84	13.74	103	91.4	0.0	159	4.5	2.5
-304			3369	13.69	12.19	75	74.3	0.0	273	5.5	3.5

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TEST B704. PROGENY PERFORMANCE UNDER RHIZOMANIA AND SBCN, IMPERIAL VALLEY, 2003-2004

(cont.)

Variety	Resistance	Description	Acre Yield			Beets/ 100'	Clean Beets	Bolting	NO3-N		Appear
			Sugar Lbs	Beets Tons	Sucrose %	No.	%	%	ppm	score	Mean
S₁ progenies from popn-921 (cont.)											
3921 -305	Rz,Bvm	RZM 2921⊗	4616	19.74	11.69	133	88.6	0.0	363	6.0	2.3
-306			7865	27.53	14.35	142	94.7	44.0	297	5.5	1.3
Mean			5081.0	18.55	13.77	149.0	88.8	2.2	281.0	5.3	2.4
LSD (.05)			2944.3	10.91	1.75	42.8	11.2	12.2	165.7	1.1	1.4
C.V. (%)			29.1	29.55	6.39	14.4	6.3	276.9	29.6	10.7	28.2
F value			2.5NS	2.212NS	4.18**	1.2NS	2.7**	2.2**	3.4**	2.3NS	2.7**

NOTES: There appeared to be a large Row 1 (rows 43-46) vs. Row 2 (rows 47-50) effect. Probably the field rows 47-50 were outside of the area with sugarbeet in 2002 and had relative low disease & SBCN severity. One rep of each entry was in each set of rows.

Test B704 with 2 reps was to screen wide differences in performance and disease reaction.. Based on these data and similar tests at Salinas under Nondiseased rhizomania, & SBCN/rhizomania evaluation, specific progenies will be selected and entered into the greenhouse screening program for resistance to SBCN.

APPEARANCE SCORE: Appearance score = canopy beauty score prior to harvest, where 1 = best and 5 = worst. 1 = estimate of how canopy (size, color, vigor, wilting, Chlorosis, necrosis, survival, etc.) would look in the absence of disease (rhizomania & SBCN). 5 = plants stunted, dead, dying and in very poor general health. 3 = approximately how lines with only Rz1 factor would rate.

DESCRIPTIONS: Y75 is broadly based Salinas gp with about 10% Bvm gp from R22. PX = paircrossed in greenhouse under paper bags. P37/8 released as CP07 has Bvm gp from WB242 and segregates for PM:pmpm. Partially inbred line C927-4 (=1927-4, 2727-4, 3927-4) was derived from one S₁ progeny (one S₀ plnat) that showed strong performance in Imperial Valley in this type of trials. C927-4 has Bvm gp from R22 (C50,C51). It may have resistance or partial resistance to SBCN. The best of these Sₙ progenies will be evaluated in container tests at Salinas in 2004-5.

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CURLY TOP EVALUATION, SALINAS ENTRIES, BSDF NURSERY
KIMBERLY, ID, 2004
DICKARD FIELD

Planted: June 7, 2004
BCT Inoculated: July 14, 2004 at 1 leaf hopper/plant

224 entries* x 3 reps, 2-row plots, 13 ft. long, sequential
70 entries* x 3 reps, 1-row plots, 13 ft. long, sequential

Variety	Description		BSDF 1st Rating 8/16/04	BSDF 2nd Rating 8/30/04	BSDF 3rd Rating 9/13/04
Hybrids					
US H11	resistant check, 10/14/02		3.0	3.0	3.3
WS-PM21	resistant check, 4/03		2.3	3.0	3.0
Phoenix	9/12/03		3.7	4.0	4.7
Beta 4776R	8/21/03		4.0	4.0	5.0
Beta 4430R	8/21/03		3.7	4.0	4.0
Monohikari	susc. ck., 1/21/03		4.3	4.7	5.7
HH142	9/12/03		3.7	4.0	4.3
Beta 4001R	8/25/03		4.0	3.7	4.3
HM-E17	3/21/02		4.0	4.0	5.3
Angelina	2/25/04		2.7	3.3	4.0
Roberta	2/25/04		3.3	3.7	4.0
US H11	resistant check, 10/14/02		2.7	3.0	3.7
Hybrids with C833-5CMS tester					
R378H5	C833-5HO	x RZM R178 (C78)	2.7	3.3	3.3
R378-6H5		x R178-6	3.3	3.7	3.7
R380-21H5		x R180-21	3.3	3.7	4.0
Y368-8H5		x Y168-8	3.3	3.7	4.3
R381-22H5	C833-5HO	x R181-22, (C81-22)	3.3	3.3	4.3
P318-6H5		x P118-6, (CP08)	3.3	4.3	4.3
Y367-5H5		x Y167-5	3.3	4.0	4.7
3931-56H5		x 1931-56	3.7	4.0	4.3
Z325-14H5	C833-5HO	x Z131-14	3.3	4.0	4.7
3931H5		x 2931, (CR931)	3.7	4.0	4.7
3941H5		x 2941, (CR941)	3.3	4.0	4.0
Monohikari	susc. check, 1/21/03		4.3	5.0	6.3
US H11	resist. check, 10/14/02		3.0	3.3	3.7
Z325H5	C833-5HO	x Z225, (CZ25/2)	3.7	3.7	3.7
CR311H5		x CR211, (CR11)	3.7	3.7	4.0
3942H5		x 2942	3.7	3.7	4.0
P207/8H5	C833-5CMS	x P007/8, (CP07)	3.3	3.7	3.7
R280/2-9H5		x R080/2-9	3.3	3.7	3.7
2930-19H5		x 1930-19 (C930-19)	3.7	4.0	4.0
2930-35H5		x 1930-35 (C930-35)	3.3	4.0	4.0

Variety	Description		BSDF 1st Rating 8/16/04	BSDF 2nd Rating 8/30/04	BSDF 3rd Rating 9/13/04
Topcross hybrids with C78/3					
R378H3	97-C562HO	x R178, (C78)	3.0	3.0	3.0
R378H5	C833-5HO	x R178	3.3	3.3	3.7
R378H50	C790-15CMS	x R178	3.3	3.3	3.7
R378H73	02-FC124HO	x R178	2.7	3.0	3.0
R378H74	02-FC1015HO	x R178	3.0	3.0	3.7
R378H77	1833-5-8HO	x R178	3.0	3.3	3.3
R378H78	1833-5-11HO	x R178	3.0	3.0	3.3
R378H59	2869-15H5	x R178	3.7	3.7	4.0
R378H60	2840-9H5	x R178	3.0	3.3	3.3
R378H62	2835-8H5	x R178	3.3	3.3	3.3
R378H63	2835-10H5	x R178	4.0	4.3	4.3
R378H64	2835-24H5	x R178	3.3	3.7	3.7
R378H66	2836-13H5	x R178	3.7	4.0	4.0
R378H67	0837-6H5	x R178	3.3	3.3	4.0
R378H68	2848-1H5	x R178	3.0	3.0	3.7
R378H80	2810-17H5	x R178	3.0	3.0	3.3
R378H81	2810-19H5	x R178	3.0	3.0	3.0
R378H42	2842HO(C842CMS)	x R178	3.3	3.0	3.3
R378H92	2790H5	x R178	3.3	3.3	3.7
Monohikari	susc. check, 1/21/03		4.3	5.0	5.7
US H11	resist. check, 10/14/02		3.0	3.3	3.3
Hybrids with SBCN resistance (Bp)					
R378H93	N265-31HO	x R178	3.0	4.0	4.0
R378H94	N265-9HO	x R178	3.0	3.7	4.3
R378H99	N265(C)HO	x R178	3.3	3.7	4.0
N324H5	C833-5HO	x RZM N224(g)	3.3	4.0	4.0
N325H50	C790-15CMS	x RZM N224(C)(g)	3.3	3.7	4.0
3931H3	C562HO	x 2931, (C931)	3.0	3.7	3.7
US H11	resist. check, 10/14/02		3.0	3.0	3.3
Testcross hybrids with C790-15CMS					
R378H50	C790-15CMS	x R178, (C78/2)	3.3	3.3	4.0
Y391H50		x RZM-ER-% Y191	3.7	4.0	4.0
Y392H50		x RZM Y292	3.7	4.0	4.0
Y375H50		x RZM Y275	3.3	3.7	4.0
R321H50	C790-15CMS	x RZM R221	3.0	3.7	4.0
R378-6H50		x R178-6	3.0	3.0	3.0
R380-21H50		x R180-6	3.0	3.0	3.0

Variety	Description	BSDF 1st Rating 8/16/04	BSDF 2nd Rating 8/30/04	BSDF 3rd Rating 9/13/04
Testcross hybrids with C790-15CMS (cont.)				
Y368-8H50	C790-15CMS x Y168-8	3.3	3.3	4.0
R381-22H50	C790-15CMS x R181-22, (C81-22)	3.3	3.7	4.3
P318-6H50	x P118-6, (CP08)	3.0	3.0	3.7
Y367-5H50	x Y167-5	3.3	3.7	4.0
3931-56H50	x 1931-56	3.0	3.7	4.0
Z331-14H5	C790-15CMS x Z131-14	3.3	4.0	4.3
3931H50	x 2931, (C931)	3.0	3.3	3.3
3941H50	x 2941, (C941)	3.0	3.7	3.7
Z325H50	x Z225, (CZ25/2)	3.3	4.0	4.3
CR311H50	C790-15CMS x CR211, (CR11)	3.3	3.3	4.0
3942H50	x 2942	3.7	3.7	3.7
P207/8H50(Sp)	x P007/8, (CP07)	3.0	3.7	3.7
R280/2-9H50	x R080/2-9	3.0	3.3	3.7
2930-19H50	C790-15CMS x 1930-19, (C930-19)	3.3	3.3	3.3
2930-35H50	C790-15CMS x 1930-35, (C930-35)	3.0	3.3	3.3
Monohikari	susc. check, 1/21/03	4.7	5.0	6.0
US H11	resist. check	3.0	3.7	3.7
Multigerm, O.P. lines				
03-C37	resist. check, C37	3.3	3.7	3.7
03-US75	Inc. 00-US75	2.7	3.3	3.3
03-SP22-0	susc. check, Inc. SP7622-0	3.7	3.7	4.0
02-US22/3	Inc. 97-US22/3	3.0	4.0	4.0
Z210	Inc. Polish %S(C)	4.0	4.3	5.3
01-EL0204	RZM 00-EL0204 (SR x Rz)	3.7	4.3	4.7
99-C46/2	Inc. U86-C46/2	3.7	3.7	4.0
99-C31/6	Inc. F86-31/6	4.3	4.7	5.3
Y390	Inc. Y190-#(C),C2,Syn 1	4.0	4.7	5.0
Y391	RZM-ER-% Y191	3.7	4.3	4.7
Y392	RZM Y292	3.7	4.7	4.7
Y393	Composite FS's,C1,Syn 1	4.0	4.3	4.3
Y375	RZM Y275	4.0	4.3	4.7
R321	RZM R221,(C26,C27)	4.0	4.0	4.7
Y367	RZM-ER-% Y167, (C67/2)	4.0	4.3	4.7
Y371	RZM-ER-% Y171	3.7	3.7	4.3
R343	RZM-ER-% R143	4.0	4.7	4.7
R336	RZM-ER-% R136,(C79-8)	3.3	3.7	4.0
R340	RZM-ER-% R140	3.3	3.7	4.0

CURLY TOP EVALUATION, SALINAS ENTRIES, BSDF NURSERY
KIMBERLY, ID, 2004
DICKARD FIELD

(cont.)

Variety	Description	BSDF 1st Rating 8/16/04	BSDF 2nd Rating 8/30/04	BSDF 3rd Rating 9/13/04
Multigerm, O.P. lines (cont.)				
R370	RZM-ER-% R170	3.7	3.7	4.3
R378	RZM-ER-% R178,(C78/3)	3.0	3.0	3.7
R378(Sp)	Inc. R178,(C78/3)	3.0	3.7	3.7
R380	RZM-ER-% R180,(C80/2)	3.3	3.7	4.0
Y369	RZM-ER-% Y169,(C69/2)	3.7	3.7	4.0
N372	RZM-ER-NR N172, (CN72)	3.7	4.0	4.3
N312	PMR-RZM-NR N112, (CN12)	3.7	3.7	4.3
03-C37	resist. check, Inc. C37	3.3	3.3	4.0
03-SP22-0	susc. check, Inc. SP7622-0	3.7	4.0	4.3
02-US22/3	Inc. 97-US22/3	3.0	3.0	3.0
P327	PMR-RZM P227, (CP03)	3.0	3.0	3.0
P328	PMR-RZM P228, (CP04)	3.0	3.0	3.0
P329	PMR-RZM P229, (CP05)	3.0	3.0	3.3
P330	PMR-RZM P230, (CP06)	3.0	3.3	4.0
P318-6	PMR-RZM P118-6, (CP08)	3.3	3.7	4.3
P307/8	PMR-RZM P207/8, (CP07)	3.0	3.3	3.3
R378	RZM-ER-% R178, (CP78/3)	3.0	3.3	3.7
R324/5	Inc. R824, (C79-2,-3)	3.3	3.7	3.7
R324	Inc. R724, (C79-2)	3.0	3.7	4.0
R325	Inc. R725, (C79-3)	3.0	3.7	4.0
R337	Inc. R637, (C79-9)	3.3	3.3	4.0
Increase of FS progeny				
Y367-5	RZM Y167-5	4.0	4.7	4.7
R378-6	RZM R178-6	3.7	3.7	4.0
R380-21	RZM R180-21	3.3	3.7	4.0
Y368-8	RZM Y168-8	4.0	4.3	4.7
R381-22	RZM R181-22, (C81-22)	4.0	4.3	5.0
P318-6(Sp)	Inc. P118-6, (CP08)	3.3	4.0	4.3
R280-6	Inc. R080-6	3.3	4.0	4.3
R280/2-9	Inc. R080/2-9	3.3	3.7	4.3
Y267-21	Inc. Y067-21	3.3	4.0	4.7
Y271-14	Inc. Y071-14	3.7	4.0	4.3
Y390-40	Inc. Y190-40	3.3	4.0	4.7
Y390-43	Inc. Y190-43	4.0	4.3	4.7
Y390-83	Inc. Y190-83	4.0	4.0	4.3
Y390-98	Inc. Y190-98	4.0	4.7	4.7
Y375-9	Inc. Y175-9	3.3	4.0	4.3

Variety	Description	BSDF 1st Rating 8/16/04	BSDF 2nd Rating 8/30/04	BSDF 3rd Rating 9/13/04
Increase of FS progeny (cont.)				
Y375-13	Inc. Y175-13	3.7	4.0	4.3
Y375-20	Inc. Y175-20	3.0	4.0	4.0
R376-89-4	Inc. R176-89-4	3.7	4.0	5.0
R376-89-10	Inc. R176-89-10	4.0	4.0	5.0
R376-89-5-4	RZM R176-89-4-5	4.0	4.0	4.3
03-C37	resist. check, Inc. C37	3.0	3.0	3.7
03-SP22-0	susc. check, Inc. SP7622-0	4.0	4.3	5.3
Multigerm, S^f,Aa populations				
3931	RZM 2931,1931aa x A, (C931)	3.7	4.0	4.3
3941	RZM 2941,1941aa x A, (C941)	3.7	4.0	4.3
3942	RZM 2942aa x A	3.7	4.0	4.7
3943	2943(C)aa x A	4.0	4.0	4.7
Z325	RZM Z225, Z125(C)aa x A,(CZ25/2)	4.0	4.3	5.0
CR311	CR111, CR111(C)aa x A, (CR11)	4.0	4.3	5.0
Multigerm, S^f,Aa SBCN populations				
N312	PMR-RZM-NR N112 (A,aa), (CN12)	4.0	4.3	4.7
N372	RZM-ER-NR N172 (A,aa), (CN72)	4.3	4.7	5.3
N324	RZM N224(g)(A,aa)	3.7	4.3	4.3
N325	RZM N224(C)(g)(A,aa)	3.7	4.0	4.3
Multigerm, S^f,Aa S_1 progeny increases				
Z131-18	Inc. Z931-18 (A,aa)	4.0	4.0	4.3
Z331-14	RZM Z131-14aa x A	4.0	4.0	4.3
Z325-105	Inc. Z125-105 (A,aa)	3.0	4.0	4.0
Z325-109	Inc. Z125-109 (A,aa)	4.0	4.7	5.0
Z325-53	Inc. Z125-53 (A,aa)	4.0	4.3	5.0
Z329-9	RZM Z225-9 (A,aa), (CZ25-9)	3.3	4.7	4.7
3931-56	RZM 1931-56aa x A	4.0	4.0	4.3
3931-120	Inc. 1931-120 (A,aa)	3.7	4.0	4.3
2941-20	Inc. 0941-20 (A,aa)	4.3	4.7	4.7
3941-107	Inc. 1941-107 (A,aa)	4.0	4.0	4.0
3941-112	Inc. 1941-112 (A,aa)	4.3	4.3	4.7
2933-14	Inc. 0933-14 (A,aa)	3.7	4.0	4.0
3933-107	Inc. 1933-107 (A,aa)	4.3	5.0	5.3
3933-113	Inc. 1933-113 (A,aa)	3.7	4.0	4.0
3933-118	Inc. 1933-118 (A,aa)	3.3	4.0	4.0
CR311-6	Inc. CR111-6 (A,aa)	3.7	3.7	4.0

Variety	Description	BSDF 1st Rating 8/16/04	BSDF 2nd Rating 8/30/04	BSDF 3rd Rating 9/13/04
Multigerm, S^f,Aa S_1 progeny increases (cont.)				
CR311-41	Inc. CR111-41 (A,aa)	3.3	3.7	4.3
CR311-88	Inc. CR111-88 (A,aa)	3.7	4.0	4.3
CR310-14-2	RZM CR111-14-2 (A,aa)	3.0	3.7	3.7
03-FC1030-15	Inc. 01-FC1030-15 (A,aa)	3.3	3.7	4.0
03-FC1030-16	Inc. 01-FC1030-16 (A,aa)	3.3	3.7	4.0
3927-4	RZM 2927-4 (A,aa), (C927-4)	3.7	4.3	4.7
03-SP22-0	susc. check, Inc. SP7622-0	3.3	4.7	5.0
03-C37	resist. check., Inc. C37	3.3	3.3	3.3
Monogerm populations and lines				
1869(C)	RZM 0869-#(C)mmaa x A, (C869)	3.0	3.7	4.0
2790	0790mmaa x A, (C790)	3.3	3.7	3.7
2842	RZM 1842mmaa x A, (C842)	3.3	3.7	3.7
2848	RZM, T-O 1848-#(C)mmaa x A	3.0	3.7	3.7
3849m	RZM 2251-2255(C)mmaa x A	3.3	3.7	4.3
3842	RZM 2842(C)mmaa x A, (C842)	3.0	3.7	4.0
3842H5	C833-5HO x 2842(C)	3.0	3.3	4.0
3842H50	C790-15CMS x 2842(C)	3.3	3.7	4.0
3869	1869(C)mmaa x A, (C869)	3.7	4.3	5.0
3869H5	C833-5HO x 1869(C)	3.7	4.0	4.3
3869H50	C790-15CMS x 1869(C)	3.3	4.0	4.0
3812	Inc. 6812M(A,aa),(C890-2,-3)	3.0	3.7	4.0
3819m	Inc. 6819 (A,aa),(C890-9,WB151)	3.3	4.0	4.0
03-FC124	RZM 02-FC124mmaa x A	3.0	3.3	3.7
03-FC124HO	02-FC124HO x " "	3.0	3.3	4.0
03-FC1015	RZM 02-FC1015mmaa x A	3.3	4.0	4.3
03-FC1015HO	02-FC1015HO x RZM 02-FC1015mmaa x A	3.3	3.7	4.3
2835	RZM,T-O 1835-#(C)mmaa x A	3.0	3.3	3.7
2836	RZM,T-O 1836-#(C)mmaa x A	3.3	3.7	4.3
2837	RZM,T-O 1836H7-#(C)mmaa x A	3.3	4.0	4.3
02-C790-15CMS	99-C790-68CMS x C790-15	3.7	3.7	4.0
02-C790-15	Inc. 00-C790-15	3.3	3.3	3.7
99-C790-68	Inc. U88-C790-68	3.3	3.7	3.7
2833-5(Sp)	RZM,T-O 1833-5-#(C)mmaa x A	3.3	3.7	4.0
2833-5NB(Iso)	NB-RZM-% 0833-5 (A,aa)	3.3	3.7	4.0
0546	Inc. 97-C546	3.0	3.3	3.3
0562	Inc. 97-C562	3.0	3.0	3.0
0762-17	Inc. 6762-17, (C762-17)	2.7	3.0	3.0

(cont.)

Variety	Description	BSDF 1st Rating 8/16/04	BSDF 2nd Rating 8/30/04	BSDF 3rd Rating 9/13/04
Monogerm populations and lines (cont.)				
3869-24	Inc. 1869-24 (A,aa)	2.7	3.0	3.7
3869-27	Inc. 1869-27 (A,aa)	3.0	3.0	3.3
3869-30	Inc. 1869-30 (A,aa)	3.0	3.0	3.7
3837-6	RZM,T-O 2837-6-#(C)	3.3	3.7	4.0
03-FC123-31	Inc. 01-FC123-31 (A,aa)	3.0	3.0	3.0
03-FC1014-22	Inc. 01-FC1014-22 (A,aa)	3.0	3.3	3.0
03-C37	resist. check, Inc. C37	3.3	3.0	3.3
0762-17	Inc. 6762-17, (C762-17)	2.7	3.0	3.0
Monogerm, SBCN resistant lines				
N365-9HOM	RZM N265-9HO x RZM N265-9(g)	3.0	3.7	4.3
N365-31HO	RZM N265-31HO x RZM N265-31(g)	3.3	3.3	3.7
N369	RZM N269-#(C)(g)(A,aa)	4.0	4.0	3.7
N365	RZM N265(g)(A,aa)	4.0	3.7	4.3
N367	RZM N267(g)(A,aa)	3.7	3.7	4.0
N366	RZM N265(C)(g)	3.3	3.3	3.3
3842	RZM 2842(C)mmaa x A, (C842)	3.0	3.0	3.0
3869	1869(C)mmaa x A, (C869)	3.0	3.3	3.0

24 entries x 3 reps, 1-row plots, 13 ft. long, sequential

S_1 progeny from popn 2843 = RZM-%S 0841H7

Variety	Description	BSDF 1st Rating 8/16/04	BSDF 2nd Rating 8/30/04	BSDF 3rd Rating 9/13/04
2843	RZM-% 0841H7(A,aa)	3.0	3.3	3.0
3843 - 1	RZM 2843mm⊗	3.0	3.7	3.7
- 2		3.3	3.3	4.0
- 4		3.7	4.0	4.3
- 6		3.3	3.7	4.3
- 8		3.3	3.7	4.3
- 9		3.0	3.3	3.3
-10		3.0	3.3	3.3
-11		3.3	4.0	3.7
-13		3.7	4.0	4.0
-16		3.3	4.0	4.3
-18		3.7	4.0	4.0
-19		3.5	3.5	4.0
-20		3.7	4.0	4.0
-22		3.3	4.0	4.3
-25		3.3	3.7	4.0

Variety	Description	BSDF 1st Rating 8/16/04	BSDF 2nd Rating 8/30/04	BSDF 3rd Rating 9/13/04

24 entries x 3 reps, 1-row plots, 13 ft. long, sequential (cont.)

S_1 progeny from popn 2843 = RZM-%S 0841H7 (cont.)

Variety	Description	BSDF 1st	BSDF 2nd	BSDF 3rd
3843 -28	RZM 2843mm⊗	3.0	3.7	3.7
-30		3.3	3.7	4.0
-32		3.3	4.3	4.3
2843	RZM-% 0841H7(A,aa)	3.0	3.7	3.7

S_1 progeny from popn 2846 = RZM-%S 0841H69

Variety	Description	BSDF 1st	BSDF 2nd	BSDF 3rd
2846	RZM-% 0841H69(A,aa)	3.0	3.0	3.3
3846 - 2	RZM 2846mm⊗	3.0	3.0	3.0
- 4		3.0	3.0	3.0
- 5		3.0	3.3	3.3
3846 - 9	RZM 2849mm⊗	3.0	3.0	3.0
-10		3.0	3.0	3.0
-12		3.0	3.0	3.0
-13		3.0	3.0	3.0
-14		3.0	3.3	3.3
-17		3.0	3.0	3.3
-18		3.0	3.0	3.0
-19		3.0	3.0	3.0
-21		3.3	3.3	3.3
2846	RZM-% 0841H69 (A,aa)	3.3	3.7	3.3

S_1 progeny from popn 2845 = RZM-%S 0841H35

Variety	Description	BSDF 1st	BSDF 2nd	BSDF 3rd
3845 - 1	RZM 2845mm⊗	3.3	3.7	3.7
- 4		3.0	3.3	3.3
- 6		3.0	3.3	3.3
- 7		3.0	3.3	3.3

S_1 progeny from popn 2790H7 = C833-5aa x C790

Variety	Description	BSDF 1st	BSDF 2nd	BSDF 3rd
2833-5(Sp)	RZM,T-O 1833-5-#(C)mmaa x A	3.3	3.3	4.0
2790	0790mmaa x A, (C790)	3.0	3.3	3.3
3891 - 1	2790H7mm⊗	3.0	3.3	3.7
- 2		3.0	3.7	4.0
- 3		3.5	4.0	4.0
- 4		3.0	3.7	3.7
- 5		3.3	4.3	4.0
- 6		3.7	4.0	3.7
- 8		3.3	4.3	4.3

(cont.)

Variety	Description	BSDF 1st Rating 8/16/04	BSDF 2nd Rating 8/30/04	BSDF 3rd Rating 9/13/04
24 entries x 3 reps, 1-row plots, 13 ft. long, sequential (cont.)				
S_1 progeny from popn 2790H7 = C833-5aa x C790 (cont.)				
3891 - 9	2790H7mm⊗	3.3	4.0	4.3
-10		4.0	4.5	4.5
-11		3.3	3.7	4.3
-12		3.3	3.7	3.7
-13		3.7	3.7	4.0
-14		3.0	3.7	4.0
-15		3.3	4.0	4.0
-16		3.0	3.7	4.0
-17		3.0	3.7	3.3
-18	2790H7mm⊗	3.0	3.7	4.3
2790	0790mmaa x A, (C790)	3.0	3.3	3.7
2835 -24	Inc. 9835-24 (A,aa)	3.0	3.0	3.0
3835 -24-1	RZM 2835-24mm⊗	3.0	3.0	3.0
-24-2		3.0	3.3	3.0
-24-3		3.0	3.3	3.3
-24-4		3.0	3.3	3.0
-24-5		3.0	3.3	3.3
-24-6		3.3	3.3	3.3
-24-7		3.0	3.0	3.0
-24-8		3.0	3.0	3.0
0762 -17	Inc. 6762-17, (C762-17)	3.0	3.3	3.0
03-SP22-0	susc. check, Inc. SP7622-0	3.7	4.3	4.7
03-C37	resist. check, Inc. C37	3.0	3.7	3.3

USDA-SALINAS ENTRIES IN FORT COLLINS DISEASE NURSERIES, FORT COLLINS, CO, 2004

For CLS:　　28 entries x 3 reps, RCB, 2-row plots, 14 ft.
For Rhizoc: 14 entries x 5 reps, RCB, 5-reps, 14 ft.

Variety	Description	Leaf Spot			Rhizoctonia		
		10Sept	17Sept	Mean	DI	%H	%0-3
Checks							
Beta 4430R	8/21/03	5.3	5.0	5.4			
Monohikari	1/21/03	3.3	3.7	3.0			
03-SP22-0	Inc. 01-SP22-0	3.0	2.7	2.8			
Multigerm, O.P.							
Y390	Inc. Y190-#(C),C2,Syn 1	4.0	3.3	3.6			
Y391	RZM-ER-% Y191	4.7	3.7	4.2			
Y392	RZM Y292	4.3	3.7	4.0			
Y393	Composite FSs,C1,Syn 1	3.7	3.3	3.4			
Y375	RZM Y275	3.7	3.7	3.5			
R321	RZM R221	4.0	3.7	3.7	2.7	28	77
Multigerm populations							
CR311	RZM CR211,111,111(C)aa x A, (CR11)	3.0	2.3	2.5			
3941	RZM 2941,1941aa x A, (C941)	4.3	3.3	3.9			
2933	RZM-% 9933 (A,aa)	3.7	2.7	3.3	3.5	11	50
Multigerm progeny lines							
3933-107	Inc. 1933-107 (A,aa)	3.7	3.0	3.3	3.4	18	56
3933-113	Inc. 1933-113 (A,aa)	3.3	3.0	2.9	3.4	10	52
3933-118	Inc. 1933-118 (A,aa)	5.0	3.5	4.1	3.4	11	62
2933-14	Inc. 0933-14	3.0	2.3	2.2	3.7	8	43
03-FC1030-15	Inc. 01-FC1030-15	4.0	3.3	3.3	2.0	49	88
03-FC1030-16	Inc. 01-FC1030-16	4.3	3.7	3.5	1.7	52	97
Fort Collins							
CR311-6	Inc. CR111- 6 (A,aa)	2.7	2.3	2.2			
CR311-41	Inc. CR111-41 (A,aa)	3.0	2.0	2.5			
CR311-88	Inc. CR111-88 (A,aa)	3.0	2.3	2.8			
CR310-14-2	RZM CR110-14-2 (A,aa)	3.5	3.0	2.9			
Monogerm populations							
03-FC124	RZM 02-FC124mmaa x A	4.0	3.0	3.4	2.8	9	75
03-FC1015	RZM 02-FC1015mmaa x A	4.3	3.3	3.7	2.4	22	85

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USDA-SALINAS ENTRIES IN FORT COLLINS DISEASE NURSERIES, FORT COLLINS, CO, 2004

(cont.)

Variety	Description	Leaf Spot			Rhizoctonia		
		10Sept	17Sept	Mean	DI	%H	%0-3
monogerm lines							
03-FC1014-22	Inc. 01-FC1014-22 (A,aa)	3.7	2.7	2.8	2.2	37	89
03-FC123-31	Inc. 01-FC123-31 (A,aa)	4.0	3.3	3.6	3.7	9	48
Hybrids							
R378H74	02-1015HO x R178	3.7	3.3	3.4	2.2	41	83
R378H73	02-124HO x R178	3.0	3.0	2.8	2.9	23	70
LSS	931002	4.7	4.0	4.3			
LSR	82105H2	3.0	2.0	2.2			
Trial mean		3.7	3.1	3.2			
Rhizoctonia Checks							
Susc. ck.	Susc.ck				2.5	31	82
Highly resist.ck.	Highly resist. ck.				1.6	49	100
Resist. ck.	Resist. ck.				1.6	62	98
Experiment mean					2.7	28	74
LSD (.05)		1.24	0.97	-.-	0.77	--	--

Cercospora scored from 0 to 10(dead). LSS = SP351069-0. LSR = (FC504CMS x FC502/2) x SP6322-0.
Mean is average of ratings on 27 Aug., 3 Sept., 10 Sept., and 17 Sept. 2004.

Rhizoctonia root rot was scored from 0(healthy) to 7(dead). DI = average rot per root. %H = percentage
of healthy roots with classes 1 and 2 considered healthy. %0-3 = % of roots likely to be taken for
processing (classes 0-3 combined).

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Variety	Description	Cercospora LeafSpot		Aphanomyces	Root Aphids	
		17 Aug	Mean	% Root Rot	Mean	%Resistant
Comm.ck.-1	Monohikari, susc. ck.	5.3	3.5	2.75	1.75	75
Comm.ck.-2	Beta 4430R, resistant ck.	7.0	4.5	2.00	3.40	7
03-SP22-0	Inc. 01-SP22-0	4.3	2.8	1.00		
03-FC1014-22	Inc. 01-FC1014-22	4.7	3.2	4.00	2.19	69
03-FC123-31	Inc. 01-FC123-31	4.3	3.0	4.00	3.00	27
03-FC124	RZM 02-FC124	6.0	3.8	4.50	3.00	20
03-FC1015	RZM 02-FC1015	5.7	3.6	4.25	3.33	17
03-FC1030-15	Inc. 01-FC1030-15	4.7	3.2	4.00	2.00	62
03-FC1030-16	Inc. 01-FC1030-16	5.3	3.3	3.25	3.00	23
2933	RZM-% 9933	5.7	3.5	3.25	3.50	13
CR311	RZM CR211aa x A	5.0	3.1	3.25	3.00	33
CR311-6	Inc. CR111-6	3.0	2.1	3.00		
CR311-41	Inc. CR111-41	4.7	2.9	4.25		
CR311-88	Inc. CR111-88	4.0	2.8	3.25		
CR310-14-2	RZM CR110-14-2	3.0	2.4	5.25		
2933-14	Inc. 0933-14	4.7	2.9	5.00	2.27	67
2933-17	Inc. 0933-17	6.7	4.0	4.50	1.57	79
3933-107	Inc. 1933-107	7.0	4.0	3.00	3.50	6
3933-113	Inc. 1933-113	5.0	3.3	4.50	2.93	27
3933-118	Inc. 1933-118	5.7	3.7	4.25	2.69	38
Y391	RZM-ER-% Y191	5.7	3.5	2.75		
Y392	RZM Y292	6.0	3.8	3.75		
Y375	RZM Y275	5.3	3.3	3.25		
R321	R221	5.7	3.6	2.50		
CR Resist 1		2.7	1.6	1.75		
CR Resist 2		3.7	2.7	7.75		
Mod CR Resist 1		5.0	3.3	6.00		
CR Susc.		7.7	4.0	5.75		
Mod CR Resist 2		4.3	3.1	8.50		
CR Resist 3		3.3	2.2	1.50		
LSD (.05)		0.85	0.50	1.06	-.--	--

NOTES: Tests managed and scored by J.Miller and M.Rekoske, Betaseed. Cercospora test at Rosemount, MN. was scored 27 July, 4 Aug, 10 Aug, and 17 Aug on the KWS scale. APH test at Shakopee, MN. was rated on a scale of 0 to 9 where 9 = dead. Root aphids rated at Shakopee in greenhouse test. 16 plants/entry rated from 1 to 4 where 1 = no aphids and 1 & 2 considered resistant.

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TEST 4104. EVALUATION OF PLANT INTRODUCTIONS (PI's), SALINAS, CA, 2004, BLOCK 2

48 entries x 4 reps, sequential
1-row plots, 11 ft. long

Planted: May 3, 2004
Harvested: December 6, 2004

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Soluble Solids %	Bolt %	Stand Count No.	Harv Count No.	DI %	%R (0-1) %	%R (0-3) %	Bolt Tend (0-3) Score	Plant Type Score
Checks													
US H11	susc.ck., 10/14/02	11565	32.05	18.05	21.48	0.0	24	23	1.8	56.9	94.4	2	3
R039	Inc. R539,(C39R)	13165	33.66	19.58	23.30	0.0	21	20	2.2	32.4	84.2	2	3
03-C37	susc.ck., Inc. U86-C37	9251	25.20	18.42	22.73	0.0	22	21	3.0	18.7	63.4	2	3
R336	RZM-ER-% R136,(C79-8)	11620	32.25	18.00	23.05	0.0	21	21	2.7	20.2	72.1	2	3
Y367	RZM-ER-% Y167,(C67/2)	13053	32.86	19.85	24.33	0.0	22	21	2.4	28.5	81.0	2	3
Y375	RZM Y275	13232	34.47	19.17	23.35	0.0	22	22	2.8	25.5	68.2	2	3
02-US22/3	Inc. 97-US22/3	9173	25.00	18.43	22.58	0.0	22	23	3.4	9.8	58.5	2	3
R321	RZM R221,(C26 x C27)	13351	36.08	18.50	22.85	0.0	21	20	2.3	39.3	80.6	2	3
Beta 4001R	resist.ck., 8/25/03	14700	37.49	19.60	23.58	0.0	24	23	1.9	48.1	93.5	2	3
Roberta	susc.ck., 2/25/04	12947	37.49	17.27	20.85	0.0	21	21	1.5	62.9	96.4	2	3
Angelina	resist.ck., 2/25/04	13080	34.67	18.85	23.28	0.0	22	22	1.8	49.1	94.0	2	3
HM-E17	susc.ck., 3/21/02	10821	28.42	18.98	23.10	0.0	24	24	3.2	7.3	63.8	2	3
Y390	Inc. Y190-#(C)	15369	39.96	19.23	23.45	13.8	23	23	2.7	30.4	73.4	2	3
P318-6(Sp)	Inc. P118-6,(CP08)	12959	35.15	18.45	22.75	16.7	21	21	3.7	9.8	44.9	2	3
P207/8(Sp)	Inc. P007/8,(CP07)	12710	33.66	18.88	23.15	23.9	22	21	2.8	18.0	72.3	2	3
N312	PMR-RZM-NR N112	14146	38.70	18.25	22.92	19.0	20	22	2.7	31.0	73.1	2	3
Beta vulgaris subsp. maritima (PI's)													
(9)PI504216	Italy, wild beet				14.90	50.7	17	11	2.9	23.4	82.3	1	5
(1)PI504226	Italy, wild beet				19.80	97.2	11	13	1.6	62.8	92.0	1	5
(11)PI504236	Italy, wild beet				17.93	82.5	21	20	1.9	45.3	93.6	1	5
(12)PI504241	Italy, wild beet				17.90	85.1	14	14	1.7	56.0	91.9	1	5
(8)PI504247	Italy, wild beet				18.60	95.6	11	10	2.1	40.3	86.1	1	5
(2)PI504250	Italy, wild beet				16.27	53.6	17	7	2.0	30.8	91.7	1	5
(13)PI504251	Italy, wild beet				19.95	58.4	11	11	1.5	59.1	98.4	1	5
(14)PI504254	Italy, wild beet				19.50	79.7	12	12	1.5	56.0	97.9	1	5

TEST 4104. EVALUATION OF PLANT INTRODUCTIONS (PI's), SALINAS, CA, 2004, BLOCK 2

(cont.)

Variety	Description	Acre Yield Sugar Lbs	Acre Yield Beets Tons	Sucrose %	Soluble Solids %	Bolt %	Stand Count No.	Harv Count No.	DI %	%R (0-1) %	%R (0-3) %	Bolt Tend Score	Plant Type Score
Beta vulgaris subsp. *maritima* (PI's) (cont.)													
(15)PI504255	Italy, wild beet				18.35	64.3	17	16	1.4	78.7	97.1	1	5
(7)PI504260	Italy, wild beet				20.88	100.0	2	5	1.7	43.9	96.4	1	5
(16)PI504275	France, wild beet	1286	5.85	10.95	13.60	77.3	19	17	3.5	0.0	59.5	1	5
(17)PI504282	France, wild beet	1050	3.16	16.92	20.20	93.5	16	16	1.2	78.6	100.0	1	5
(6)PI518324	UK, En IDBBNR 5818	2725	8.71	15.82	21.68	85.0	20	21	2.5	18.5	79.8	1	5
(18)PI518338	UK, En IDBBNR 5832	2730	8.67	15.77	22.38	76.6	18	15	2.0	34.9	94.5	1	5
(10)PI518398	Ireland, IDBBNR 5892	5162	13.91	18.60	22.48	64.2	21	19	1.3	72.0	100.0	1	5
(5)PI518405	Ireland, IDBBNR 5899	5171	13.91	18.52	22.15	73.1	20	19	1.3	71.2	100.0	1	5
(19)PI518421	UK, En IDBBNR 5915	4623	15.08	15.50	22.63	58.6	18	17	3.3	7.8	66.4	1	5
(20)PI518436	UK, En IDBBNR 5930	2719	8.06	16.83	22.28	56.8	20	20	1.9	41.8	97.2	1	5
(21)PI540594	France, WB 848	2449	8.06	15.48	21.55	68.2	19	20	1.6	53.6	98.9	1	5
(4)PI540608	France, WB 862	2786	8.18	17.30	23.27	95.9	15	15	1.3	72.5	100.0	1	5
(22)PI540616	France, WB 870	2802	7.86	17.83	24.03	72.4	21	19	1.6	53.3	97.5	1	5
(23)PI540617	France, WB 871	2646	8.06	16.40	23.10	85.5	20	20	1.6	55.2	97.7	1	5
(3)PI540619	France, WB 873	2829	9.07	15.63	22.30	88.3	22	22	1.5	59.5	99.0	1	5
(24)PI540620	France, WB 874	2085	7.06	14.80	20.40	93.1	21	22	1.7	40.2	98.8	1	5
(25)PI540629	UK, WB 883	2133	6.25	17.35	22.55	83.0	21	19	1.8	35.5	98.7	1	5
Checks													
03-US75	Inc. 00-US75,susc.ck.	10667	30.24	17.63	21.25	0.0	23	22	2.9	19.2	73.0	2	3
03-SP22-0	Inc. 01-SP22-0,susc.ck.	8555	23.79	17.98	21.15	0.0	24	22	3.3	15.7	56.0	2	3
Y391	RZM-ER-% Y191	14399	36.69	19.60	23.55	0.0	22	24	3.0	15.4	68.3	2	3
R378	RZM-ER-% R178,(C78/3)	13650	35.07	19.45	23.83	0.0	23	23	2.5	34.3	77.5	2	3
P328	PMR-RZM P228,(CP04)	12846	34.07	18.85	22.90	0.0	23	22	3.1	13.2	61.8	2	3
P330	PMR-RZM P230,(CP06)	14252	37.90	18.80	22.88	0.0	22	22	2.5	30.2	78.5	2	3
R324/5	Inc. R824,(C79-2,-3; WB41,42)	9998	26.81	18.65	23.15	0.0	21	20	2.8	17.4	71.9	2	3

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TEST 4104. EVALUATION OF PLANT INTRODUCTIONS (PI's), SALINAS, CA, 2004, BLOCK 2

(cont.)

Variety Description	Acre Yield Sugar Lbs	Beets Tons	Sucrose %	Soluble Solids %	Bolt %	Stand Count No.	Harv Count No.	DI %	%R (0-1) %	%R (0-3) %	Bolt Tend Score	Plant Type Score
Mean	8650.2	23.52	17.74	21.5	42.4	19.3	18.7	2.2	38.0	83.7	1.5	4.0
LSD (.05)	1575.5	4.06	1.60	1.9	31.0	4.4	3.4	0.7	21.1	16.7	0.0	0.0
C.V. (%)	13.0	12.31	6.42	6.3	52.2	16.5	13.1	21.5	39.7	14.2	0.0	0.0
F value	79.0**	77.16**	9.47**	12.3**	13.2**	7.2**	13.3*	8.4**	7.7**	6.4**	-.-	-.-

NOTES: Rhizomania was very mild. Based known resistant and susceptible checks, rhizomania data are not reliable and should not be used. Roots from wild beets from Italy bolted too young to evaluate %S and sugar yield. Harvested roots were weighed and tested for %soluble solids.

SUGAR BEET RESEARCH
USDA-ARS Sugarbeet Research Unit in Fort Collins, Colorado

2004 REPORT
Section B

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USDA-ARS-NPA Sugar Beet Research Unit's Mission Statement

Utilize distinctive site environmental and disease-free characteristics and specifically developed team expertise to: develop new knowledge and adapt biotechnologies to modify host-pathogen relations that affect disease resistance, pathogenesis, and epidemiology in sugar beet and other plant species pertinent to sugar beet cultivation; discover new information and techniques to identify and produce genotypes exhibiting superior disease and stress tolerance and agronomic qualities; and provide new knowledge that improves production efficiency and biochemical processing characteristics of sugar beet.

(Projects 420, 421, 440, 441, 443, 903, and 904)

CONTENTS

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UNITED STATES DEPARTMENT OF AGRICULTURE
AGRICULTURAL RESEARCH SERVICE
WASHINGTON, DC

AND

BEET SUGAR DEVELOPMENT FOUNDATION
DENVER, COLORADO

NOTICE OF RELEASE OF FC720, FC722, and FC722CMS MONOGERM SUGARBEET
GERMPLASM

The USDA Agricultural Research Service (ARS), in cooperation with the Beet Sugar Development Foundation (BSDF), announces the release of FC720, FC722, and FC722CMS (PI 636335, PI 636336, and PI 636337, respectively) sugarbeet germplasm. These germplasm were developed in the breeding program of Drs. L. Panellla and L. E. Hanson, USDA-ARS, Fort Collins, Colorado. FC720 has good resistance to root-rotting strains (AG-2-2) of *Rhizoctonia solani* Kühn and good to moderate resistance to cercospora leaf spot caused by *Cercospora beticola* Sacc., but is not resistant to the *Beet curly top virus* (BCTV). FC722 has good resistance to root-rotting strains (AG-2-2) of *Rhizoctonia solani* and good to moderate resistance to cercospora leaf spot caused by *Cercospora beticola*, but is BCTV susceptible. FC720 and FC722 are populations from which to select rhizoctonia and cercospora resistant, monogerm, O-type parents to infuse some rhizoctonia and leaf spot resistance on the female side of hybrids, and FC722CMS provides a CMS female with these characteristics. FC720 is released from seed production 20001017, FC722 from 19961010HO, and FC722CMS from 19961010HO1.

FC720 is an O-type germplasm with 73% green (rr) hypocotyls (26 plants counted) and is segregating for monogerm (mm) and self-sterility (S^s). FC722 is an O-type germplasm with 15% green hypocotyls (rr) (59 plants counted) and is segregating for monogerm (mm) and self-sterility (S^s). Both germplasm have FC708 (PI 590845), a rhizoctonia- and cercospora-resistant monogerm O-type release from the Fort Collins program as one parent. The other parent, C718 (PI 590849), is a germplasm released from the USDA-ARS sugarbeet improvement program in Salinas, CA, and is moderately susceptible to virus yellows, moderately resistant to bolting, and moderately resistant to BCTV; it has very good combining ability for root and gross sugar yield.

FC720 is a product of six generations of mass selection for rhizoctonia resistance among $R\_$ (pink hypocotyl) progeny in the seed harvested from C718 (rr) of the cross combination (C718 (rr)//(C718/FC708)) $R\_$. The initial population had 25% of its genes from FC708 and 75% from C718. The smallest population size was 13 plants. FC722 also is a product of six generations of cyclic mass selection for rhizoctonia root rot resistance. The source population was the cross C718/FC708; and the initial population had 50% of its genes from FC708 and 50% from C718. The smallest population size was 13 plants. FC722CMS is the genetic-cytoplasmic male sterile

equivalent of FC722 backcrossed nine times. The original cross was C718CMS/FC708. It was backcrossed continually to the populations, from which FC722 was derived, and went through five generations of cyclic mass selection for rhizoctonia root rot resistance.

FC720, FC722, and FC722CMS exhibited good resistance to rhizoctonia root rot when tested under strong disease pressure. FC720, FC722, and FC722CMS's performance was equal to the rhizoctonia-resistant check in disease index (DI) ratings (DI of 0 = no root rot and 7 = all plants dead), except in 1999 when FC722CMS was significantly less resistant than the resistant check (FC703), but significantly more resistant than the susceptible check (FC901/C817). FC720, FC722, and FC722CMS always performed significantly better than the susceptible check. FC720 had mean disease indices (DI's) of 4.1, 4.0, and 1.7 (1999-2001, respectively), whereas the resistant check had mean DI's of 3.8, 3.8, and 2.6. FC722 had mean DI's of 4.0, 4.2, and 2.4; and FC722CMS had mean DI's of 4.6, 4.2, and 2.4. Percentages of resistant plants (those rated 0 or 1) were 8, 3, and 47 for FC720; 6, 0, and 17 for FC722; 2, 0, and 13 for FC722CMS; 22, 13, and 98 for the highly resistant check and 12, 3, and 21 for the resistant check (FC703) (1999-2001, respectively).

FC720, FC722, and FC722CMS also exhibited resistance to cercospora leaf spot when tested in an artificial epiphytotic. In three years of tests (1998 1999, 2002), they were significantly better than the susceptible check, and not significantly different from the resistant check, except for FC722, which had significantly less resistance than the resistant check in 1999. The following DI ratings (DI of 0 = no leaf spot and 10 = all plants dead) represent the most severe rating (last of three or four ratings each season). The DIs of FC720, FC722, and FC722CMS, respectively, were 3.2, 3.7, and 3.7; 3.8, 3.7 and 4.0; 3.7, 3.7 and 4.0; DIs of the resistant check (FC504CMS/FC502-2//SP6322-0) were 2.8, 2.7 and 3.7; DIs of the susceptible check (SP351069-0) were 5.8, 6.3, and 5.0 (in 1998, 1999, and 2002, respectively). FC720, FC722, and FC722CMS did not show tolerance to the BCTV, even though the parent line, C718, was BCTV-resistant.

In 2002, FC720, FC722, and FC722CMS were planted on May 3 in one-row plots, replicated six times at the USDA-ARS Fort Collins Research Farm. Plots were 3.04 m long with 56 cm between rows and 20 to 25 cm within-row spacing. Roots were harvested on October 8 and sent to the Western Sugar Co. tare lab in Scotts Bluff, NE for analyses. The average sucrose concentration and sugar loss to molasses of three commercial varieties – Beta 6045, HM1955, Monohikari – was used as a standard for comparison. Sucrose concentrations of FC720, FC722, and FC722CMS, respectively, were 86.0%, 70.2%, and 76.3% of the standard, and in sugar loss to molasses, FC720, FC722, and FC722CMS were 113.3%, 114.9% and 123.7%, respectively, of the standard.

Breeder seed of FC720, FC722, and FC722CMS is maintained by USDA-ARS and will be provided in quantities sufficient for reproduction upon written request to Sugarbeet Research, USDA-ARS, Crops Research Laboratory, 1701 Center Ave., Fort Collins, CO 80526-2083. Genetic material of this release will be deposited in the National Plant Germplasm System where it will be available for research purposes, including development and commercialization of new varieties/cultivars. We request that appropriate recognition be made of the source when this germplasm contributes to a new cultivar. U.S. plant variety protection will not be requested for

FC720, FC722, or FC722CMS.

BSDF Project 903 – Evaluation of Contributed Lines for Resistance to *Rhizoctonia solani*, a Causal Fungus of Sugar Beet Root Rot.

L.E. Hanson and L. Panella

Annually, for over thirty years, the sugar beet breeding program in Fort Collins has included the production of an artificial epiphytotic through inoculation with *Rhizoctonia solani* to evaluate and select for resistance to root rot caused by this pathogen. We have been pleased to participate and lead this cooperative research project between the ARS, Colorado State University, and the BSDF.

In 2004 the project involved field studies conducted at the Crops Research Lab-Fort Collins Research Farm near Wellington, CO. Randomized, complete-block designs with five replicates were used to evaluate ARS breeding germplasm and Plant Introduction accessions. *Rhizoctonia*-resistant line FC703, highly resistant FC705-1, and susceptible FC901/C817 were included as internal controls.

One-row plots, planted May 17th, were 14 feet long with 22 inches between rows and 8-10 inches within-row spacing. The field was sprayed twice with Betamix Progress, Upbeet, and Stinger (June 21 and 26) to control weeds. The field was thinned by hand and irrigated as necessary. Inoculation with dry, ground, barley-grain inoculum of *Rhizoctonia solani* AG2-2 isolate R-9 was performed on July 27th; immediately after inoculation, a cultivation was performed so as to throw soil into the beet crowns. Beets were harvested September 13 through 17. Each root was rated for rot on a scale of 0 to 7 (dead) as previously described. ANOVAs were performed on disease indices (DIs), percent healthy roots (classes 0 and 1 combined), and percentage of roots in classes 0 thru 3. Percentages were transformed to arcsin-square roots to normalize the data for analyses. LSDs are provided for comparing entries with those of our internal checks.

The moderate daytime temperatures in the summer of 2004 (Figure 1), combined with a moderate inoculum load, contributed to a moderate root rot epidemic. Severe disease developed by mid-September. Mean DIs across all tests for highly resistant FC705-1, resistant FC703, and susceptible FC901/C817 controls were 2.1, 2.4, and 3.9 respectively. Mean DIs for these controls in 2003 were 3.2, 3.3 and 5.5 respectively. Percentages of healthy roots were 36.1, 31.1, and 12.3% for these controls. Percentages of roots in disease classes zero thru three were 88.9, 80.1, and 37.7% respectively. The highest and lowest DIs for the evaluated lines were 6.7 and 1.5, respectively.
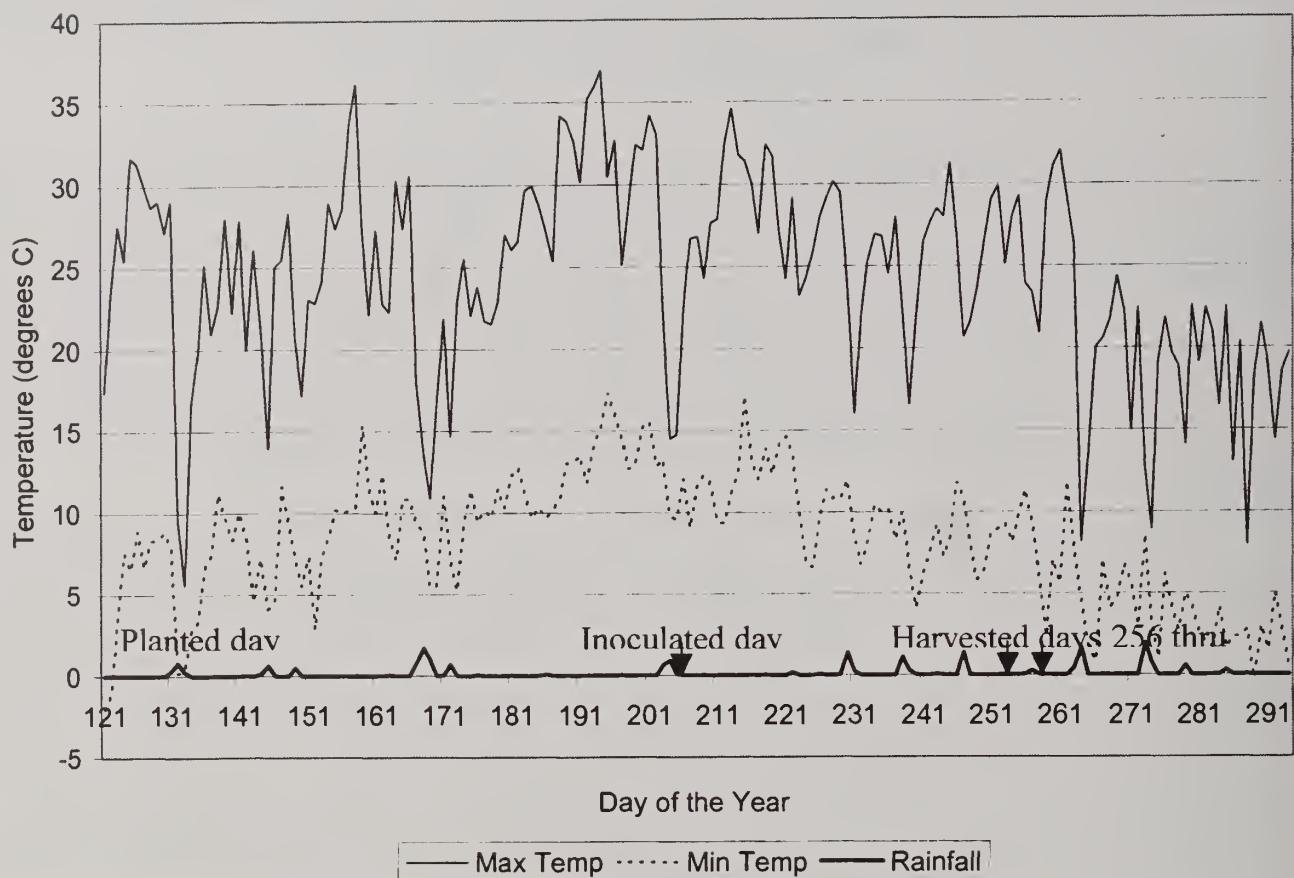
Figure 1. Summary of the weather data for 2004 Rhizoctonia root rot nursery.

Table 1. Summary data of the 2004 Rhizoctonia root rot nursery.
The experiment mean, the mean of the susceptible check, the mean of the resistant check, and the mean of the highly resistant check are given for each of the experiments in the nursery. LSD is at the t=0.05 level.

	Disease Index				Percent Healthy (classes 0&1)					Percent in Classes 0 to 3				
Exp.	Mean	Sus.	Res.	H. Res.	LSD	Mean	Sus.	Res.	H. Res.	LSD	Mean	Sus.	Res.	H. Res.	LSD
1R	3.7	4.2	2.5	2.2	0.8	2.4	0.0	7.4	17.8	9.1	45.1	31.8	91.2	97.6	22.3
2R	5.0	3.3	2.6	2.5	1.2	1.1	9.4	12.6	13.8	6.6	24.0	56.0	87.2	89.2	26.5
3R	3.0	3.1	1.9	2.1	0.9	24.1	25.0	47.4	40.4	14.0	64.7	51.0	89.4	87.6	15.8
4R	3.0	3.4	2.4	1.5	0.9	29.7	18.4	45.2	62.4	15.9	58.3	43.8	74.8	98.4	18.1
5R	3.1	4.6	2.7	2.2	0.9	24.4	3.8	25.8	30.4	16.5	59.7	19.6	70.6	85.0	19.2
7R	2.7	2.5	1.6	1.6	0.8	27.6	30.6	52.0	49.4	14.4	73.9	66.1	97.8	100.0	17.5
8R	3.1	3.9	2.4	2.3	0.7	16.2	9.6	35.8	31.0	14.3	58.6	38.4	76.0	87.4	16.2
9R	3.2	4.8	2.7	1.8	0.8	16.8	9.0	19.6	51.8	17.6	60.4	20.2	76.8	93.4	18.4
10R	4.3	4.8	3.2	3.3	0.7	7.9	7.0	22.4	17.0	12.9	22.1	12.0	54.0	51.8	15.2
11R	3.6	3.9	2.2	1.8	0.8	15.0	10.4	42.8	47.2	17.0	47.5	38.0	83.4	98.2	16.8

Percent in Classes is the transformed value (arcsin-square root)
Mean = Experiment Mean;
Sus. = Susceptible Check (FC901/C817);
Res. = Resistant Check (FC703);
H Res. = Highly Resistant Check (FC705/1)

B8

BSDF Project 904 – Evaluation of Contributed Lines for Resistance to *Cercospora beticola*, Causal Fungus of Cercospora Leaf Spot.

L.E. Hanson and L. Panella

The breeding program in Fort Collins has created an annual artificial epiphytotic through inoculation with *Cercospora beticola* for over forty years. This epiphytotic has been used to evaluate and select for resistance to leaf spot caused by *C. beticola*. We have been pleased to participate in and lead this cooperative research project between the ARS, Colorado State University, and the BSDF.

In 2004 the project primarily involved field studies conducted at the Irrigation Research Center near Yuma, CO. Randomized complete-block designs, with three replications, were used to evaluate commercial and experimental entries. Internal controls included a highly susceptible synthetic (SP351069-0) and a resistant check (FC504CMS/FC502-2//SP6322-0). Two-row plots were 12 feet long, with 22-inch row spacing and an 8- to 10-inch within-row plant spacing. The trial was planted on April 21. Hail and winds hit the plots in early May and caused severe stand losses. Inoculations of remaining plots were performed on July 8 and July 22. Hail hit the field five times in 2004. Only three of the original seven experimental plots had enough plants to rate. Evaluations were made on August 27, September 3, 10, and 17, with the peak of the epidemic occurring between the second and third ratings. The field was sprayed with Nortron (April 26), Betamix Progress (May 31, June 9, and June 30), Upbeet (May 31 and June 9), and Stinger (June 9) to control weeds. The field was irrigated as necessary.

The moderate temperatures in the summer of 2004 and low moisture (Figure 2) contributed to a moderate leaf spot epidemic, which did not become severe enough to rate until the end of August. Disease severity increased through the first two weeks of September. By the third rating (September 10), means of the resistant and susceptible internal control were 3.0 and 4.3 (scale of 0-10), respectively across the nursery. In 2003 (September 12) these means were 3.5 and 5.8, respectively. Means of contributor lines in 2004 ranged from 2.7 to 5.3. Table 2 shows the data for the nursery from the three ratings in September.
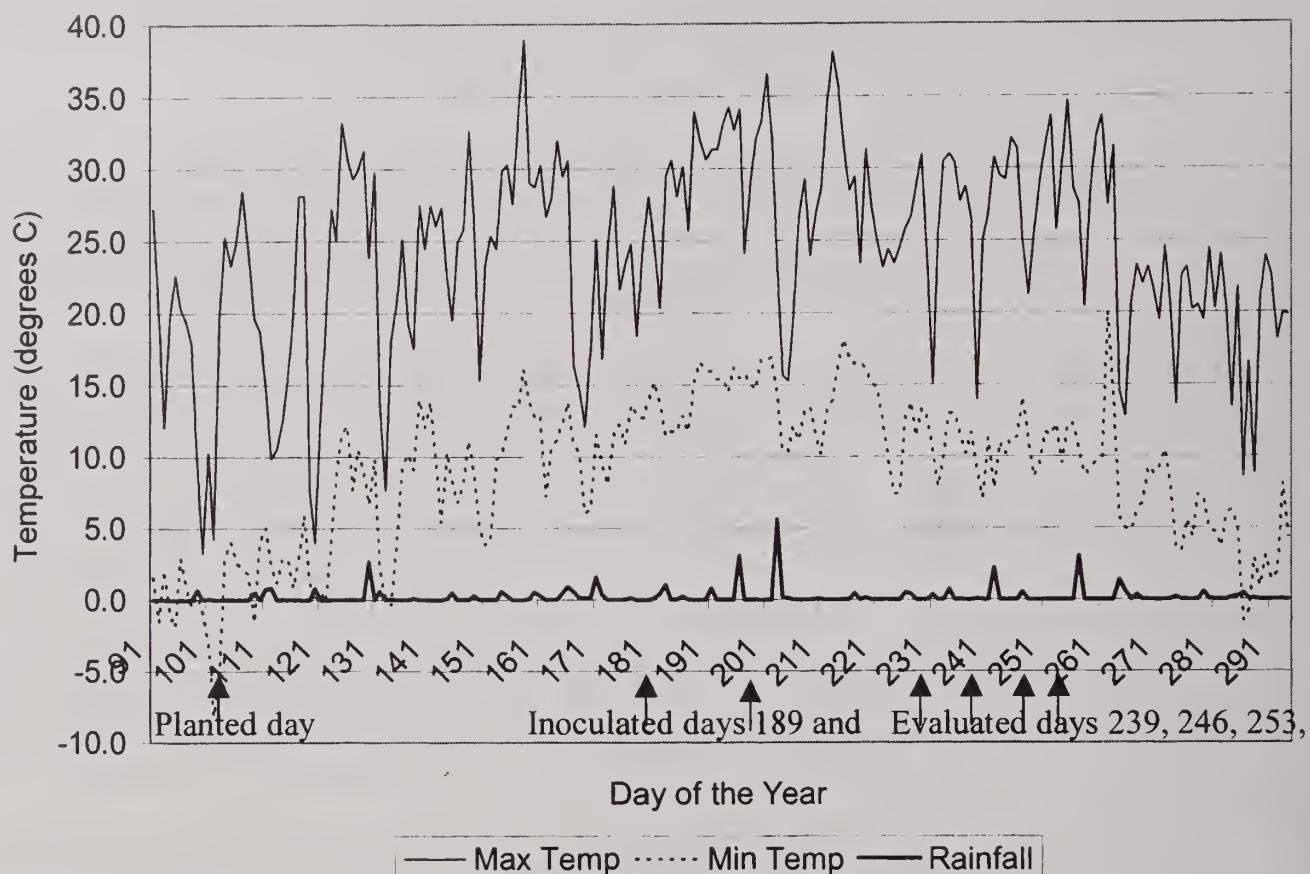
Figure 2. Summary the 2004 weather data for Cercospora Leaf Spot Nursery.

Table 2. Summary data of the 2004 Cercospora leaf spot disease nursery.
The experiment mean, the mean of the susceptible check, and the mean of the resistant check are given for each of the experiments in the nursery, for each evaluation date.

Exp.	September 5th Disease Index				September 12th Disease Index				September 19th Disease Index			
	Mean	Sus.[1]	Res.[2]	LSD	Mean	Sus.	Res.	LSD	Mean	Sus.	Res.	LSD
1A	3.6	3.5	2.3	1.08	3.9	4.0	3.0	1.18	3.2	3.0	2.3	1.13
3A	3.0	4.2	1.3	0.82	3.4	4.3	3.0	0.96	2.8	3.7	2.3	0.84
4A	3.4	4.3	2.7	1.35	3.7	4.7	3.0	1.24	3.1	4.0	2.0	0.97
Mean	3.33	4.00	2.10		3.67	4.33	3.00		3.03	3.57	2.20	

[1]Cercospora Susceptible Check - SP351069-0
[2]Cercospora Resistant Check - FC 504CMS/FC 502-2//SP6322-0

BSDF Project 420 – Screening Biological Control Agents for *Rhizoctonia solani* Control on Sugar Beets.

L.E. Hanson, L. Panella, A.L. Hill, G.M. Preston

Rhizoctonia root and crown rot (caused by the fungus *Rhizoctonia solani* Kühn) is the most common and most serious fungal root disease of sugar beet in the United States. The disease is endemic in beet producing areas of the United States. *Rhizoctonia solani* also causes a damping-off in sugar beet seedlings. If the infection is light, the fungus may cause crown rot or dry rot canker on maturing roots later in the season. Thus control of this fungus in the seedling stage might offer some reduction in disease later in the season, as well as improving crop stands.

Biological control can provide an alternative to chemical pesticides which are the subject of increasing regulation and restrictions due to environmental and public health concerns. Biological control is compatible with host genetic resistance and thus can be used in an IPM program. While resistance to *R. solani* is available, it does not provide complete immunity and resistance is not well expressed in seedlings, thus the addition of other control methods is desirable.

In 2003, four *Pseudomonas fluorescens* strains (PMS382, F113, SBW25, and ΔWSP) from G. M. Preston were used. All four strains showed biological control activity against *Pythium ultimum* in Dr. Preston's work. *Trichoderma virens* strains included two strains (G-6 and G-4) from Texas cotton field soil with activity against damping-off in cotton, two UV-mutants of strain G-6, one (AB1-5) with biological control activity on cotton and one (AB1-4) without biological control activity, and isolates obtained from sugar beet, SB-1, T-4, T-5, and T-33. In addition, two *T. koningii* strains (Tk-7 and TkG-12), one *T. longibrachiatum* and one *T. atroviride* strain were used in tests. Additional strains from sugar beet are being obtained and will be included in future tests.

In *in vitro* antibiosis tests against *R. solani*, all four bacterial isolates inhibited *R. solani* growth on potato dextrose agar (PDA). In tests with *Trichoderma*, isolate PMS382 inhibited the growth of all strains of *Trichoderma* tested. The three other *P. fluorescens* strains did not significantly inhibit growth of any of the *T. virens* strains, indicating that these bacterial and fungal strains may be used in combination. Growth of *T. atroviride, T. longibrachiatum* and *T. koningii* was inhibited by F113, but not by SBW25 or ΔWSP. None of the *Pseudomonas* strains were significantly inhibited by any of the fungal strains. When seed was soaked in a *Pseudomonas* suspension (F113 or SBW25), air dried, and treated with *Trichoderma* grown in wheat bran+peat moss, both *Pseudomonas* and *Trichoderma* could be isolated from the seed.

In antibiosis tests against *R. solani*, *T. virens* strains G-6, T-2, T-3, T-4 T-33 and SB-1 inhibited *R. solani*, while G-4, AB1-5, LH-2 and AB1-4 showed no inhibitory activity. Strain G-6 is a "q" strain of *T. virens* that produces the antibiotic gliotoxin, which has activity against *R. solani*. Strain G-4 is a "p" stain of *T. virens* that produces the antibiotic gliovirin, which has activity against *Pythium ultimum*, but not against *R. solani*. Our results suggest that T-2, T-3, T-4, T-33, and SB-1, which we isolated from sugar beet, are "q" strains and LH-2 is a "p" strain. The *T. atroviride, T. longibrachiatum,* and *T. koningii* strain Tk-7 did not inhibit *R. solani in vitro* while *T. koningii* strain TkG12 showed weak inhibition of *R. solani* AG-4 with little effect on AG-2-2.

In greenhouse biological control assays, no significant disease control was observed with any of the *Pseudomonas* isolates. Seed treatment with wheat bran+peat moss preparations of G-6, and AB1-5 significantly increased seedling survival in all tests. Seed treatment with SB-1 and G-4 each showed significantly increased seedling survival in more than half of all tests, but survival was lower

than with G-6 and results were more variable. T-4, T-5 and T-33 showed variable activity in two or more tests, but survival was variable. No significant increase in survival was observed with AB1-4 in any tests. All of the *T. virens* strains colonized the root system well. No significant disease control was observed for the *T. atroviride*, *T. longibrachiatum*, or *T. koningii* strains.

Different application methods for the *Rhizoctonia solani* inoculum were tested in the field in 2004. In previous years, the survival in control plots (for example, 3% seedling survival in 2003) was too low, and is a very severe test for a control method. Inoculum was diluted with ground vermiculite and applied either in furrow, as a side dressing at planting, or as a side dressing one week after planting. The inoculation in furrow at planting gave the most consistent disease levels. All replicates showed some damping-off with this treatment. Side dressing with inoculum at planting gave some disease, but results were variable with some replicates showing no significant disease. Side dressing one week after planting gave seedling survival levels that were not significantly different from the vermiculite control survival. A dilution of one part *Rhizoctonia* infested ground barley to 9 parts ground vermiculite gave (15-25% seedling survival).

In field tests for biological control activity with a subset of isolates, none of the seed treatment with significantly increased seedling survival under heavy *R. solani* pressure compared to the bare seed control (Table 3). However, two isolates, AB1-5 and SB-1, gave significantly higher seedling survival than the fungicide control treatment. In addition to *Rhizoctonia*, in 2004, this field has a severe flea beetle infestation. There was a great deal of variability in stand in the different replicates, and it may be that flea beetle damage was involved.

Differences between activity in greenhouse and field tests are not unusual with biological control agents. For example, isolate G-6 was from acid soil and is reported to provide control in acid soils, but little or no control in alkaline soils. The soil in this field was approximately pH 7.6. Isolate SB-1, which gave significantly increased control in the field in 2002, did not provide significant disease treatments control in 2003 at a 95% probability, however, SB-1 and T-4 gave significantly higher survival than the untreated or fungicide controls at a 90% probability. In 2004, isolate SB-1 did not give significantly higher survival than the carrier control, but did give significantly higher survival than the fungicide control at a 95% probability. Isolate T-4 did not provide any significant disease control. Isolate AB1-5, which did not provide significant disease control in the field in 2003 or 2003, had significantly higher seedling survival in 2004 than the fungicide control. This isolate gave significant disease control in the greenhouse. At the level of infestation in our tests, a fungicide seed treatment of Apron+Thiram did not give any detectable disease control compared to bare seed in either 2003 or 2004. Seedling survival levels were not significantly different between the fungicide-treated seed and those treated with the wheat bran and peat moss carrier used for applying *Trichoderma* strains.

No growth promotion of sugar beet was detected for any of the *Trichoderma* isolates on seedlings. There were no significant differences in the timing of seed germination, seedling height, seedling fresh weight or root weight between control plants and those treated with *Trichoderma* at two or three weeks after planting in the absence of *Rhizoctonia solani*. No significant growth promotion was observed for any of the *Pseudomonas* isolates either for any of these parameters.

Table 3. Emergence and survival of sugar beet (FC403) seedlings with and without *R. solani* (AG2-2) treated with a wheat bran + peat moss preparation of *T. virens* strain G-6 or with the wheat bran + peat moss carrier alone.

Treatment	Percent survival, field[1]
Carrier control[2]	61 ab[3]
Fungicide[4]	60 ab
AB1-5	69 a
SB-1	62 ab
T-4	55 abc
T-33	38 def
TkG-12	49 bcd
T-5	45 cde
R. solani (R9)	23 fgh
Fungicide[4] + *R. solani*	13 h
AB1-5 + *R. solani*	35 defg
SB-1 + *R. solani*	35 defg
T-4 + *R. solani*	20 gh
TkG-12 + *R. solani*	29 efgh
T-5 + *R. solani*	20 gh
T-33 + *R. solani*	25 fgh

[1] Average percent seedling survival from six replicates 21 days after planting under field conditions.
[2] Carrier control is wheat bran and peat moss.
[3] Percentages in the same column followed by the same letter are not significantly different by Fischer's LSD ($\alpha=0.05$).
[4] Fungicide seed treatment was Apron (metalaxyl) and Thiram (tetramethylthiuram disulfide).

Mycoparasitic ability was tested by plating potential biocontrol agents and sugar beet pathogens on a low nutrient medium and examining the area of interaction. In some cases, interaction was not observed because growth of the pathogen was inhibited by the *Trichoderma* and only dead hyphae of the pathogen were found in the area of *Trichoderma* growth. However, when interaction occurred, hyphal coiling (Fig. 3) was observed, and hyphal penetration was detected for all strains of *T. virens* except AB1-4 and AB1-5. These two isolates previously had

been demonstrated to show no mycoparasitism *in vitro*.

Figure 3. Hyphal coiling of *Trichoderma* around *Rhizoctonia solani*. Hyphal coiling is associated with mycoparasitism in *Trichoderma*. Arrow indicates *Trichoderma* mycelium

BSDF Project 421 – Variability in *Fusarium oxysporum* from sugar beets in the United States.

L.E. Hanson, L. Panella, A.L. Hill

Fusarium yellows causes significant reduction in root yield, sucrose percentage and juice purity in affected sugar beets. Research in our laboratory and others on variability in *Fusarium oxysporum* associated with sugar beets demonstrated that isolates that are pathogenic on sugar beet can be highly variable. A better understanding of this variability is important in the efforts to test for Fusarium yellows resistance in beets and efforts to breed for resistance.

In 2002, 113 *Fusarium* isolates were obtained from sugar beets and in 2002-2003 these were identified to species. Isolates included 60 *F. oxysporum*, 19 *F. solani*, 16 *F. equiseti*, 8 *F. avenaceum*, 7 *F. acuminatum*, and one isolate each of *F. proliferatum, F. subglutinans*, and *F. verticillioides*. *Fusarium subglutinans* has been reported from stored sugar beet, but not from actively growing beets.

In 2003, 134 isolates of *Fusarium* were obtained from sugar beet and identified to species. Isolates included 76 *F. oxysporum*, 17 *F. equiseti*, 14 *F. solani*, 6 *F. culmorum*, 5 *F. acuminatum*, 3 *F. proliferatum*, 3 *F. crookwellense*, and two isolates each of *F. avenaceum, F. graminearum, F. semitectum, F. subglutinans*, and *F. verticillioides*. *Fusarium culmorum* has been reported to cause a root rot of sugar beet under drought conditions in Europe, and there were drought conditions in some areas in 2003. We did receive some root rot samples in 2003, from which *Fusarium* were obtained. *Fusarium solani* was isolated from both sugar beet and dry bean from two fields with root rot. *Fusarium solani* has been reported to cause a root rot in sugar beet, and dry or root rot of dry bean. The causal agent for dry root rot of bean is reported to be *Fusarium solani* f.sp. *phaseoli*, with some host specificity for bean. Since these root rot samples were from the same field, it needs to be determined whether the same isolates could affect both bean and beet. *Fusarium graminearum* has been associated with sugar beet in Europe and has been isolated from beets in storage. *Fusarium crookwellense* and *Fusarium semitectum* have not to our knowledge been reported from growing sugar beet. The majority of isolates in 2003 were from samples with yellowing symptoms. However, there was very little vascular discoloration in a number of these samples. Pathogenicity tests for these isolates are ongoing. At least one *Fusarium graminearum* isolates causes stunting and yellowing of beets.

In 2004, 89 isolates of *Fusarium* were obtained from diseased beets and identified. Isolates included 35 *F. oxysporum*, 15 *F. equiseti*, 6 *F. graminearum*, 4 *F. acuminatum*, 2 *F. avenaceum*, 2 *F. culmorum*, and one isolate each of *F. lateritium, F. semitectum*, and *F. sporotrichioides*. All or the *F. graminearum* isolates to date have been from samples taken in the more northern areas. Seven of the eight isolates were from Minnesota, with the other isolate from northern Wyoming. *F. sporotrichioides* has been isolated from beets in storage, but not from beets in the field.

Over the three years of this project, a total of 392 *Fusarium* isolates have been obtained, with the majority (53%) being *Fusarium oxysporum*. Of the *F. oxysporum*, approximately 25% of the isolates tested to date are pathogenic on sugar beet. In addition, isolates of at least four other *Fusarium* species have been determined to cause yellows symptoms on sugar beet. No isolates of *F. equiseti* have been found pathogenic on sugar beet in our greenhouse assay, but this species has been associated with postharvest mold problems in sugar beet (Bosch & Mirocha 1992).

In addition to isolates from sugar beet, two *F. oxysporum* f. sp. *spinaciae* (FOS) isolates were

kindly provided by Dr. L. duToit. These isolates were obtained from spinach and had been demonstrated to be pathogenic on spinach. In greenhouse tests, both spinach isolates were pathogenic on sugar beet with a moderate level of virulence. This is consistent with the three FOS isolates obtained and tested previously. While these isolates are pathogenic on sugar beet, and 10 of 12 isolates from sugar beet tested were pathogenic on spinach, preliminary genetic evidence (Fig. 4) indicates that the FOS are more similar to one another than to isolates from beet.
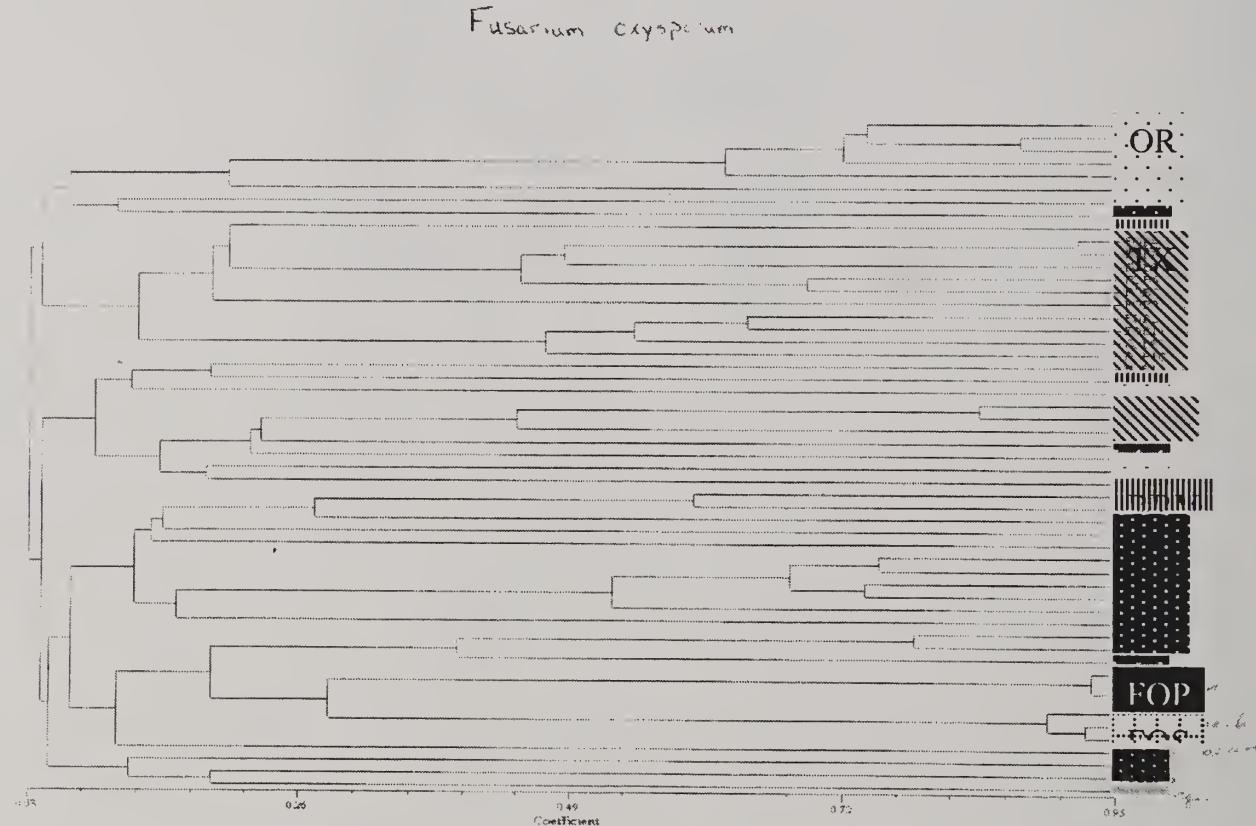
Figure 4. RAPD analysis shows a high degree of genetic variability in *Fusarium oxysporum* from sugar beet compared to isolates from spinach (FOS) or dry bean (FOP).

Isolates of *F. oxysporum* so far obtained in this study include isolates from California, Colorado, Minnesota, Montana, Nebraska, North Dakota, Oregon, Washington, and Wyoming. Pathogenic isolates identified so far are from Colorado, Montana, North Dakota, Oregon, and Washington.

DNA has been extracted from all pathogenic isolates obtained in 2000 and 2001, as well as isolates provided by collaborators and used in RAPD analysis to examine genetic variability. Pathogenic isolates have been found to be a diverse group (Fig. 4). Diversity between *F. oxysporum* isolates pathogenic on beet was much higher than that reported in some other *formae speciales*, and isolates of FOS and *F. oxysporum* f.sp. *phaseoli* examined in our study showed greater clustering than the beet isolates.

To look for differences in host response in different isolates, isolates of *F. oxysporum* from different states, and one isolate of *F. solani*, were tested for virulence on *Fusarium*-susceptible sugar beet germplasm FC716 and two beet lines with reported resistance to Fusarium yellows. When the area under the disease progress curve (AUDPC) was determined for each of these isolates, variability was found between different isolates on the different beet lines (Figure 5). This demonstrates

variability in the interaction between different *F. oxysporum* isolates and sugar beet lines. This is an indication of a probable race situation in this pathogen.
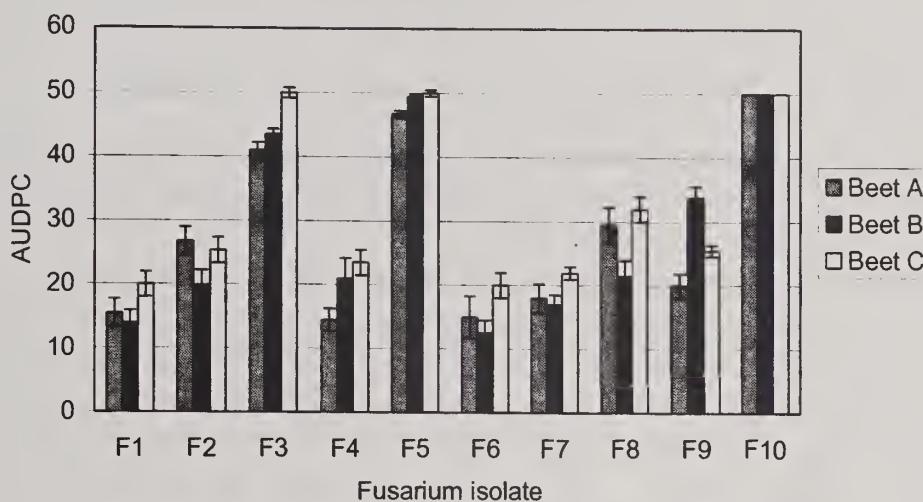
Figure 5. Area under the disease progress curve for disease severity ratings for *F. oxysporum* isolates on three different sugar beet lines, two with reported resistance to *Fusarium oxysporum* (Beet A & B) and one susceptible (Beet C = FC716).
Each point is an average from 10 plants. Isolate F1 is *F. solani*. Other isolates are *F. oxysporum*.

The finding of other species causing Fusarium yellows is of concern since current disease control measures are aimed at controlling *F. oxysporum*. Rotation with small grains and corn has been recommended for Fusarium yellows control, but *F. acuminatum*, *F. avenaceum*, *F. graminearum*, and *F. verticillioides* can be pathogens on small grains and *F. verticillioides* on corn. Thus these rotations might not aid in disease control.

The presence of several of these species of sugar beet also could be of concern for other crops grown in rotation with sugar beet, whether or not they cause disease on sugar beet. Several of the species isolated from sugar beet are generally reported to be grain pathogens. For example, *F. equiseti* was the second most commonly isolated species after *F. oxysporum*. While no isolates of this species were pathogenic on growing sugar beet, isolates of this species are important pathogens of cereal grains. Similarly, isolates of *F. avenaceum*, *F. acuminatum*, *F. culmorum*, *F. graminearum*, and *F. verticillioides* are pathogens of grains. In addition, isolates of *F. avenaceium*, *F. acuminatum*, *F. culmorum*, *F. graminearum*, and *F. solani* have been reported to cause dry rot in potatoes and *F. solani* can be a pathogen of dry beans. At this time it is not known whether the isolates from sugar beet can affect these other crops, but this could be of concern for infection of crops in the rotation.

BSDF Project 440 – Rhizoctonia Root Rot Resistance and Development of Genetic Resistance in Sugar Beet

L. Panella & L. E. Hanson
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Summary of Literature

Twenty-five years ago, Leach and Garber (1970) reviewed resistance to Rhizoctonia infection and concluded, "In general, while it has been possible to identify differences among cultivars or selections in susceptibility to Rhizoctonia infection, it is extremely rare that a high degree of resistance has been found or produced by selection or breeding within a susceptible host species." However, one of the most effective and environmentally safe ways to manage plant disease is with resistant germplasm (Sherf and MacNab, 1986). Soilborne pathogens like Rhizoctonia are often difficult to control chemically. Fumigation is expensive, providing only a temporary solution. The use of Quadris™[1] provides the first real chemical control for this disease. However, we are finding that timing of application is crucial. Additionally, spot spraying can be time consuming, and spraying a whole field because of a few patches of disease also can be expensive. The use of resistant germplasm, coupled with crop rotation and other cultural practices, can provide excellent management of diseases caused by *Rhizoctonia solani*.

In sugar beet (*Beta vulgaris* L.), Rhizoctonia root- or crown-rot is caused by *Rhizoctonia solani* (AG-2-2). Seedling damping-off in sugar beet also is caused by *R. solani* AG-4. Root-rot is endemic in sugar beet growing areas across the United States. John Gaskill began breeding for resistance in the late 1950s and released his first resistant germplasm in 1966 (Gaskill, 1968). Current Rhizoctonia resistant germplasm has a level of resistance in which there is no yield loss under disease pressure in the field (Ruppel and Hecker, 1994). It was realized early that natural field epiphytotics did not produce the necessary consistent, uniform disease pressure for recurrent mass selection (Pierson and Gaskill, 1961). Artificially induced epiphytotics (Ruppel et al., 1979; Schneider et al., 1982) were developed to provide uniform, heavy disease pressure to be able to perform mass selection or recurrent field selection (Hecker and Ruppel, 1977).

The resistance to *R. solani* in sugar beet developed by John Gaskill is polygenic, involving at least two loci, two or three alleles, and modifying genes in some populations (Hecker and Ruppel, 1975). Broad-sense heritability has been estimated at about 0.65, and there are nonadditive components of the variance (Hecker and Ruppel, 1975). In a study by Hecker and Ruppel (1976) dominance effects were present in diploid, triploid, and tetraploid resistant hybrids. Relatively high heritability has aided in the development of increasing host plant resistance to Rhizoctonia root- and crown-rot, and we have released over 15 germplasm lines in the last 10 years. Rhizoctonia Resistance has been released in O-type maintainer, CMS female, and multigerm-pollinator germplasm and remains a very important means of reducing crop damage by this disease (Herr, 1996). Genetic resistance to Rhizoctonia root rot has been an ongoing development from this project at Fort Collins. Several resistant germplasms have been released in the last five year to use as

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[1]Mention of a trademark or manufacturer by the USDA does not imply its approval to the exclusion of other products or manufacturers.

parents of hybrid cultivars or to provide source populations from which Rhizoctonia resistant parents were selected or which were crossed to provide resistant parents (Panella and Ruppel, 1996; Panella and Ruppel, 1997; Panella, 1999; Panella, 2001).

Epidemiological and control studies have been reported regularly from this project (Ruppel et al., 1988). Pathogen survival in varied crop debris and soil and the interaction of pesticides with Rhizoctonia have been reported on the literature (Ruppel, 1985; Ruppel 1991; Ruppel and Hecker, 1982; Ruppel et al., 1982). In a 3-year study, positive significant or highly significant correlations between disease severity indices and percent decreases in yield and purity parameters indicated that there were no hidden losses to Rhizoctonia root rot in our resistant germplasms (Ruppel and Hecker, 1994).

Recently, researchers attempting to determine the anastomosis group (AG) of *Rhizoctonia solani* isolates have used several new biotechnological techniques (including RFLP, RAPD, and isozyme analyses), with some notable successes in distinguishing among, and even within some, of these groups. Recently there was a report of a definitive assay to distinguish those isolates in AG-2-2 or AG-4 that cause sugar beet root rot and damping-off, respectively, from nonpathogenic isolates obtained from soil (Lubeck and Poulsen, 2001).

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Justification for this research

Rhizoctonia root rot continues to be a problem in most sugar beet-growing areas in the United States, and is a growing problem world wide. The practice of short rotations and the expansion of growing areas into infested areas compound the problem. The result is a reduction in net returns to growers as well as processing losses due to reduced sucrose and purity of rotted or partially rotted beets. Genetic resistance, coupled with judicious cultural measures, is a more economical and practical method of reducing losses caused by this fungus than is a strictly chemical control regime. There is also a strong need of combining Rhizoctonia root rot resistance with Rhizomania resistance.

This has been an ongoing and productive project, and has been the only research project with the goal of discovering, developing, and releasing Rhizoctonia-resistant germplasm to industry breeders, our major external customers. Although several relatively resistant germplasms have been

developed, we need to continue to combine this resistance with resistance to other diseases, and to develop a faster means of introgressing this resistance into more commercially acceptable materials

OBJECTIVES:

1   Develop Rhizoctonia-resistant populations from different genetic sources of resistance.

2   Plant mother roots and selections for seed production and ultimate release to breeders for use as populations from which to develop Rhizoctonia- and rhizomania-resistant parents in hybrid cultivars.

3   Combine resistance to *Rhizoctonia* with that of other important pathogens (esp. Rhizomania) in germplasm with good agronomic performance.

4   A longer range goal (in collaboration with Mitch McGrath, USDA-ARS East Lansing) is the development of molecular markers linked to the genes in sugar beet controlling resistance to *R. solani*.

Materials and Methods:

Field isolation plots and greenhouse isolation chambers in Fort Collins will be used for seed production from mother roots and selections of advanced germplasms having been field selected for resistance to Rhizoctonia root rot. The Fort Collins environment has proven extremely valuable in these efforts. The arid climate, low organic matter content of the soils, and hot, dry winds are not conducive to the development of soilborne or foliar diseases. Therefore, when artificial epiphytotics, developed by Gaskill and Ruppel, are created to test sugarbeet for resistance to Rhizoctonia root rot there is little confounding of the results by the presence of other diseases.

Selected resistant populations resulting from crosses with material containing the single *Rz* gene source of resistance to Rhizomania will be sent to Salinas for field selection for Rhizomania resistance. Alternating cycles of selection in Salinas and Fort Collins (and Kimberley, ID for curly top resistance) will be used to increase disease resistance. Seed increases will be made and the germplasms will be released as adequate seed becomes available.

Molecular genetic studies will concentrate on looking at the response of the sugar beet to attack by *Rhizoctonia solani*. A longer range goal (in collaboration with Mitch McGrath, USDA-ARS East Lansing) is the development of molecular markers linked to the genes controlling resistance to *R. solani*. Populations are being developed at East Lansing for this purpose and molecular markers (SSRs & AFLPs) at both Fort Collins and East Lansing.

2004 Field Research on Rhizoctonia Root Rot of Sugar Beet

Annually, for over thirty years, the sugar beet breeding program in Fort Collins has included the production of an artificial epiphytotic through inoculation with *Rhizoctonia solani* to evaluate and select for resistance to root rot caused by this pathogen. In 2004 the project involved field studies conducted at the Crops Research Lab-Fort Collins Research Farm near Wellington, CO.

Randomized, complete-block designs with five replicates were used to evaluate ARS breeding germplasm and Plant Introduction accessions. *Rhizoctonia*-resistant line FC703, highly resistant FC705-1, and susceptible FC901/C817 were included as internal controls. One-row plots, planted May 17th, were 14 feet long with 22 inches between rows and 8-10 inches within-row spacing. The field was sprayed twice with Betamix Progress, Upbeet, and Stinger (June 21 and 26) to control weeds. The field was thinned by hand and irrigated as necessary. Inoculation with dry, ground, barley-grain inoculum of *Rhizoctonia solani* AG2-2 isolate R-9 was performed on July 27th; immediately after inoculation, a cultivation was performed so as to throw soil into the beet crowns. Beets were harvested September 13 through 17. Each root was rated for rot on a scale of 0 to 7 (dead) as previously described. ANOVAs were performed on disease indices (DIs), percent healthy roots (classes 0 and 1 combined), and percentage of roots in classes 0 thru 3. Percentages were transformed to arcsin-square roots to normalize the data for analyses. LSDs are provided for comparing entries with those of our internal checks.

The moderate daytime temperatures in the summer of 2004 (Figure 1 – Project 903), combined with a moderate inoculum load, contributed to a moderate root rot epidemic. Severe disease developed by mid-September. Mean DIs across all tests for highly resistant FC705-1, resistant FC703, and susceptible FC901/C817 controls were 2.1, 2.4, and 3.9 respectively. Mean DIs for these controls in 2003 were 3.2, 3.3 and 5.5 respectively. Percentages of healthy roots were 36.1, 31.1, and 12.3% for these controls. Percentages of roots in disease classes zero thru three were 88.9, 80.1, and 37.7% respectively. The highest and lowest DIs for the evaluated lines were 6.7 and 1.5, respectively.

Table 4.   Allotment of Fort Collins "FC" numbers (3-digit numbers)

"FC" numbers are "convenience" numbers for "seed releases" or purposes where a permanent line designation is needed — i.e. a number that does not change from generation to generation where little or no selection pressure is applied. Initially, an "FC" no. was written thus "FC 501" [now FC727], "FC 502 CMS" [now FC715CMS], etc. Sublines (from selfing) were designated thus, "FC 502/2" [now FC709-2], "FC502/3" [now FC502-3], etc. The same applies when the line is substantially changed by selection without selfing.

100's	Early releases
200's	Rhizoctonia, rhizomania resistant, combined with other resistances
300's	Leaf Spot Resistant (LSR), combined with rhizomania resistance
400's	Parental lines and special genetic stocks
Below 500	Originally LeRoy Powers -
500's	Leaf Spot Resistant (LSR), Type-O lines & male steriles [CMS]
600's	LSR-Curly Top Resistant (CTR), type-O lines & male steriles [CMS]
700's	Rhizoctonia Resistant
800's	LSR-CTR-Rhizoctonia resistant
900's	Pollinators, LSR-CTR type

Rhizoctonia-Resistant Populations Under Development

Rhizoctonia root rot continues to be a problem in most sugar beet-growing areas in the United States, and is a growing problem world wide. The practice of short rotations and the expansion of growing areas into infested areas compound the problem. The result is a reduction in net returns to growers as well as processing losses due to reduced sucrose and purity of rotted or partially rotted beets. Genetic resistance, coupled with judicious cultural measures, is a more economical and practical method of reducing losses caused by this fungus than is a strictly chemical control regime. There is also a strong need of combining Rhizoctonia root rot resistance with Rhizomania resistance.

This has been an ongoing and productive project, and has been the only research project with the goal of discovering, developing, and releasing Rhizoctonia-resistant germplasm to industry breeders, our major external customers. Although several relatively resistant germplasms have been developed, we need to continue to combine this resistance with resistance to other diseases, uncover new sources of resistance, and work to more quickly introgress this resistance into germplasm with higher sucrose yield potential.

Current Research 2004 – Germplasm under development:

Current Rhizoctonia-resistant germplasm under development consists of populations being jointly developed with Dr. Robert Lewellen in Salinas (numbers 2 through 5 below). These populations are being improved to combine Rhizoctonia and Rhizomania resistant in a genetic background with good sucrose yield potential. Additionally, a population providing root maggot resistance along with Rhizoctonia and Cercospora resistance is under development with Larry Campbell in Fargo. Finally, potential new sources of Rhizoctonia have been identified, are being retested and will be crossed to sugar beet with high sucrose potential and Rhizomania resistance.

1   With the release of FC724 and FC710(4X) in 2003 and FC720, FC722, and FC722CMS, in 2004; after the release of FC723, and FC723CMS this fall, the germplasm remaining from the program of Dr. Richard Hecker will have been either evaluated, recombined, improved and released, or shelved.

2   Rhizoctonia root rot resistance multigerm base population developed by a cross between FC709-2 and a Salinas germplasms, 2915, which has been tested in Salinas & Fort Collins as FC1030). – includes populations 03-FC1030-15 and 03-FC1030-16 being reselected in Fort Collins for Rhizoctonia resistance.

     a.   2915 (sp) RZM 1915-#m 1913-# aa x A (Salinas); Seed harvested from aa (ms) plants open-pollinated by A- (fertile) plants. This population will segregate for A-:aa, Rz-:rzrz, $s^s s^s$:s^f-, (>½ s^f), R-:rr, It will be multigerm, have moderate to good tolerance to virus yellows, curly top, bolting, Erwinia; variable for reaction to powdery mildew, production traits. Individual plants will be either Aa or aa.

Background of population is mostly from OP, MM lines such as C46, C37.

3 20021028; FC709-2 (Fort Collins release) x 9933 (Salinas germplasm) [(2000A011 x 19921024)rr blk F$_2$]; Should segregate for rhizoctonia and cercospora leafspot resistance (FC709-2), multigermity, root aphid resistance (FC709-2 and 9933), tolerance to curly top, Virus Yellows, powdery mildew, Erwinia, rhizomania (9933) S^f-, A-, in a fertile cytoplasm – tested in Salinas as 04-FC1028.

4 Sib-lines of FC201 – 01-FC1014-22 (A,aa); 01-FC1014A; 01-FC1014H5 – C833-5 CMS x 01-FC1014A; 03-FC1015 – Rzm (C833-5 mmaa x FC1014) mmaa x A; 03-FC1015HO, CMS equivalent of the previous – (C833-5 CMS x 01-FC1014A) x Rzm (C833-5 mmaa x FC1014) mmaa x A.

5 20021022 – [2000A010aa [9931] x 20001009 [sel(FC907 x FC709-2)F3]blk increase-blk – was sent to Salinas and reselected in Fort Collins.
   a. 9931 = Advanced Base breeding population at Salinas with resistance/tolerance to Rz, CT, VY, Pm, Erwinia, bolting – segregates for Aa:aa, Sf, Multigerm.

Progress in 2004

The population (FC708/2890&2859) was released as FC201. It is a segregating population with a high frequency of the *Rz1* allele conferring resistance to rhizomania caused by *Beet necrotic yellow vein virus*. FC2201 is segregating for resistance to root-rotting strains (AG-2-2) of *Rhizoctonia solani* Kühn and to the sugarbeet root aphid (*Pemphigus* sp.), has moderate resistance to cercospora leaf spot caused by *Cercospora beticola* Sacc., to black root caused by *Aphanomyces cochlioides* Drechsl., and the *Beet curly top virus*. FC201 is a heterogeneous population from which to select disease resistant monogerm, O-type parents to infuse multiple disease resistance on the female side of hybrids. There is no CMS equivalent. FC201 is released from Salinas seed production 01-FC1014 and has been tested as 00-FC1014 and 01-FC1014.

1 Final testing and seed increase of monogerm O-type lines with and without and CMS equivalents, selected in the 1996 Rhizoctonia nursery were completed and two of those lines have been released and the rest are to be released in the fall (listed above). (Table 6 below).

2 This population (FC709-2/2915) was divided into four breeding lines selected in Fort Collins, CO, and Kimberley, ID. Two have been selected for resistance to Rhizoctonia (individual plant selections and half-sib families selections), one was selected for resistance to Rhizoctonia and curly top virus (half-sib families selections), and one was selected for resistance to curly top (half-sib families selections). Three of the populations were planted in Dr. R. Lewellen's Rhizomania/steckling nursery for selection for resistance to rhizomania (*Rz* – Holly gene source) and for agronomic performance. Selected roots will be increased for further sucrose and rhizomania testing, selection, and release.

3 Seed of 04-FC1028 was received from R.T. Lewellen in Salinas after selection for sucrose and

rhizomania and will be reselected for Rhizoctonia resistance.

4   Selections made in a (FC709-2 x FC907)F$_2$ population in the Rhizoctonia nursery were increased in the greenhouse and tested in the Rhizoctonia root rot, Cercospora leaf spot and curly top nurseries. This population will be re-selected for percent sucrose and increased for release. (Table 6 below).

5   20021022 – [2000A010aa [9931] x 20001009 [sel(FC907 x FC709-2)F3]blk increase-blk – was sent to Salinas and the parent line 20021002 was reselected for Rhizoctonia resistance in Fort Collins.

6   A number of accessions from the NPGS *Beta* collection that had shown Rhizoctonia-resistance in the Sugarbeet CGC screening program have been identified. Those PIs with seed available were re-screened in 2003. Special attention will be paid to those accessions screened in 1987 and 1992 because the tests in those years appear to have been unreliable. Crosses were not made this year, but these and other PIs are being screened (Table 5 below). Crosses will be made between any that appear to have resistance using a female parent with high sucrose yield potential and with Rhizomania resistance. The goal is to develop Rhizoctonia-resistant populations from potentially different sources of resistance, from which breeders will be able to select resistant hybrid parents or germplasm to cross into programs developing Rhizoctonia-resistant hybrid parents (See table 5 below).

Table 5. Experiment 2R, 2004 – Plant Introductions from Pullman Washington through the Sugar Beet Crop Germplasm Committee.

Entry	Seed Source	Release Description	DI[1]	% Hlthy[2]	% 0 - 3[3]	Z%[4] Hlthy	Z% 0 - 3[4]
		(<2.2 significantly better than susceptible check) LSD[5]	1.1			5.9	23.7
		CV	18.0			296.9	80.3
		Susceptible Check[6]	3.3	9	56	15.6	49.0
		Experiment Mean	5.0	1	24	1.6	23.7
		Highly Resistant Check[7]	2.5	14	89	17.1	77.5
		Resistant Check[8]	2.6	13	87	16.3	71.7
641	PI 540662	WB 916, France	4.9	0	25	0.0	27.0
642	PI 540664	WB 918, France	5.0	0	12	0.0	15.7
643	PI 540667	WB 921, France	5.8	0	5	0.0	5.7
644	PI 540689	WB 943, Belgium	4.1	0	43	0.0	41.1
645	PI 546377	Cal IDBBNR 5654, US	3.2	3	69	4.9	56.2
646	PI 504236	Wild Beet, Italy	4.7	0	13	0.0	18.1
647	PI 540620	WB 874, France	5.1	0	20	0.0	22.7
648	PI 518421	En IDBBNR 5915, UK	5.4	0	14	0.0	14.6

Table 5. Experiment 2R, 2004 – Plant Introductions from Pullman Washington through the Sugar Beet Crop Germplasm Committee.

Entry	Seed Source	Release Description	DI[1]	% Hlthy[2]	% 0 - 3[3]	Z%[4] Hlthy	Z% 0 - 3[4]
		(<2.2 significantly better than susceptible check) LSD[5]	1.1			5.9	23.7
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		Experiment Mean	5.0	1	24	1.6	23.7
		Highly Resistant Check[7]	2.5	14	89	17.1	77.5
		Resistant Check[8]	2.6	13	87	16.3	71.7
649	PI 518338	En IDBBNR 5832, UK	4.8	0	22	0.0	24.1
650	PI 540660	WB 914, France	5.8	0	3	0.0	4.4
651	PI 504241	Wild Beet, Italy	5.6	0	17	0.0	18.4
652	PI 504226	Wild Beet, Italy	5.3	0	7	0.0	7.0
653	PI 518436	En IDBBNR 5930, UK	4.8	0	16	0.0	18.2
654	Ames 19156	IDBBNR 9555, Russian Federation	3.1	6	68	9.3	58.9
655	PI 504201	Wild Beet, Italy	4.8	0	33	0.0	31.2
656	PI 504250	Wild Beet, Italy	5.7	0	8	0.0	8.8
657	PI 504251	Wild Beet, Italy	5.3	0	18	0.0	17.9
658	PI 504260	Wild Beet, Italy	6.7	0	0	0.0	0.0
659	PI 518303	En IDBBNR 5797, UK	5.6	0	17	0.0	16.0
660	PI 518324	En IDBBNR 5818, UK	5.7	0	0	0.0	0.0
661	PI 558505	IDBBNR 5892, Ireland	6.1	0	8	0.0	12.8
662	PI 518405	IDBBNR 5899, Ireland	5.7	0	16	0.0	20.6
663	PI 540594	WB 848, France	5.4	0	7	0.0	11.7
664	PI 540636	WB 890, France	5.1	0	17	0.0	18.7
665	PI 540658	WB 912, France	5.7	0	18	0.0	22.2
666	PI 504215		4.9	0	19	0.0	23.4
667	PI 504212		4.5	2	44	3.9	44.2
668	PI 504227		5.4	0	13	0.0	13.6
669	PI 504224		4.3	0	26	0.0	30.1
670	PI 504222		4.5	3	24	5.3	25.1
671	PI 504218		4.6	0	37	0.0	36.8
672	PI 504229		5.3	0	10	0.0	11.9
673	PI 504231		4.6	0	16	0.0	15.5
674	PI 504187		4.4	0	42	0.0	39.7
675	PI 504249		5.5	0	17	0.0	13.7
676	PI 504246		6.1	0	4	0.0	5.3
677	PI 504245		5.6	0	20	0.0	18.0
678	PI 518344		4.8	0	20	0.0	23.1
679	PI 518336		5.7	0	5	0.0	6.6

Table 5. Experiment 2R, 2004 – Plant Introductions from Pullman Washington through the Sugar Beet Crop Germplasm Committee.

Entry	Seed Source	Release Description	DI[1]	% Hlthy[2]	% 0 - 3[3]	Z%[4] Hlthy	Z% 0 - 3[4]
		(<2.2 significantly better than susceptible check) LSD[5]	1.1			5.9	23.7
		CV	18.0			296.9	80.3
		Susceptible Check[6]	3.3	9	56	15.6	49.0
		Experiment Mean	5.0	1	24	1.6	23.7
		Highly Resistant Check[7]	2.5	14	89	17.1	77.5
		Resistant Check[8]	2.6	13	87	16.3	71.7
680	PI 504252		4.7	0	30	0.0	27.0
681	PI 518317		6.0	0	4	0.0	5.3
682	PI 518357		5.0	0	31	0.0	30.2
683	PI 504244		6.0	0	13	0.0	11.0

[1]Disease Index is based on a scale of 0 (=healthy) to 7 (= plant dead).
[2]Percent of healthy roots (disease classes 0 and 1 combined).
[3]Percent of diseased roots likely to be taken for processing (disease classes 0 through 3 combined).
[4]Percentages were transformed to arcsin-square roots to normalize the data for analyzes.
[5]P=0.05; There were missing plots, however LSD was estimated as if all plots were present.
[6]FC901/C817 - susceptible check
[7]FC705/1 - highly resistant check

Table 6. Experiment 5R, 2004 – Fort Collins Breeding Germplasm.

Seed Source	Release Description	DI[1]	% Hlthy[2]	% 0 - 3[3]	Z%[4] Hlthy	Z% 0 - 3[4]
	(<3.7 significantly better than susceptible check) LSD[5]	0.9			14.8	17.2
	CV	22.4			45.5	26.0
	Susceptible Check[6]	4.6	4	20	7.1	22.9
	Experiment Mean	3.1	24	60	25.9	52.6
	Highly Resistant Check[7]	2.2	30	85	32.3	75.0
	Resistant Check[8]	2.7	26	71	29.9	58.2
19921019	FC729 – FC712/A4, 3 cycles Rhizoc, MM	2.7	28	69	31.2	57.1
19951016HO	FC723 – EL44/FC708 mm	2.4	32	76	34.1	63.8
19951016HO1	FC723CMS – EL44/FC708 CMS	2.1	46	79	42.0	68.5
19961010HO1	FC722CMS – C718/FC708CMS	2.8	23	71	27.9	60.6
19961014	FC724 New Release 2003	2.4	40	78	38.5	63.6
19961015	FC720 -- C718/(C718/FC708)	1.9	47	88	43.4	74.3
20021011H	FC709-2	2.4	28	80	30.7	69.2

Table 6. Experiment 5R, 2004 – Fort Collins Breeding Germplasm.

Seed Source	Release Description	DI[1]	% Hlthy[2]	% 0 - 3[3]	Z%[4] Hlthy	Z% 0 - 3[4]
(<3.7 significantly better than susceptible check) LSD[5]		0.9			14.8	17.2
CV		22.4			45.5	26.0
Susceptible Check[6]		4.6	4	20	7.1	22.9
Experiment Mean		3.1	24	60	25.9	52.6
Highly Resistant Check[7]		2.2	30	85	32.3	75.0
Resistant Check[8]		2.7	26	71	29.9	58.2
20001022	FC710(4X)	2.1	33	93	34.7	77.9
20011007	F3 (907 x 709-2) for RhzcR - hs 10A-1775	2.4	30	86	29.9	68.4
20011060	[FC712 x 9931(Salinas)] F2	5.1	0	12	0.0	14.1
20021001H	LSR EL & FC polycross	2.8	34	67	34.6	59.3
20021002	RhzcR/mR - (FC907 x FC709-2) x 9931	4.4	7	17	7.0	18.2
20021018HO	FC712/Mono-Hy A4	1.9	44	92	41.4	75.4
20021018HO1	FC712/MonoHy A4 CMS	2.5	33	74	34.6	62.2
20021028	FC709-2 x 9933 (root aphid resistant)	4.8	4	18	6.7	21.9
2003A033	FC201	3.1	25	64	28.6	56.6
2003A034	FC301	4.5	3	25	6.1	26.5
20031019	RhizocR x RhizomR; FC712 x 9931(Salinas)	3.5	17	50	21.4	44.9
20031020	RhcR MM pop; 2915aa x FC709-2	2.8	29	68	28.0	62.1
2003A035	01-FC1014A	4.0	11	37	16.8	37.2
2003A036	01-FC1014H5 - C833-5 CMS x 01-FC1014A	4.6	3	23	6.1	24.7
2003A037	01-FC1014-22 (A,aa)	3.2	27	52	28.3	46.6
2003A048	03-FC1015 - Rzm (C833-5 mmaa x FC1014) mmaa x A	3.2	20	57	25.7	49.7
2003A049	03-FC1015HO – CMS equivalent of above	4.6	3	26	4.6	29.2
2004A008	EL51	1.8	54	88	47.9	77.3

[1]Disease Index is based on a scale of 0 (=healthy) to 7 (= plant dead).
[2]Percent of healthy roots (disease classes 0 and 1 combined).
[3]Percent of diseased roots likely to be taken for processing (disease classes 0 through 3 combined).
[4]Percentages were transformed to arcsin-square roots to normalize the data for analyzes.
[5]P=0.05; There were missing plots, however LSD was estimated as if all plots were present.
[6]FC901/C817 - susceptible check
[7]FC705/1 - highly resistant check

Table 7. Experiment 7R, 2004 – USDA-ARS Salinas, California germplasm and joint Salinas-Fort Collins Germplasm.

Entry	Seed Source	Release Description	DI[1]	% Hlthy[2]	% 0 - 3[3]	Z%[4] Hlthy	Z% 0 - 3[4]
		LSD[5]	0.8			14.4	17.5
		CV	22.7			38.8	21.7
		Susceptible Check[6]	2.5	31	82	33.2	66.1
		Experiment Mean	2.7	28	74	29.3	63.6
		Highly Resistant Check[7]	1.6	49	100	44.6	90.0
		Resistant Check[8]	1.6	62	98	52.0	86.1
781	R321	RZM R221; lso12	2.7	28	77	30.8	64.9
782	3933	RZM-% 9933 (A,aa); lso59	3.5	11	50	15.4	45.8
783	3933-107	Inc. 1933-107(A,aa); lso26	3.4	18	56	24.2	48.7
784	3933-113	Inc. 1933-113 (A,aa); lso27	3.4	10	52	14.3	49.1
785	3933-118	Inc. 1933-118(A,aa); lso28	3.4	11	62	14.4	52.3
786	2933-14	Inc. 0933-14; lso41	3.7	8	43	16.0	41.0
787	03-FC1030-15	Inc. 01-FC1030-15; lso39	2.0	49	88	44.6	72.8
788	03-FC1030-16	Inc. 01-FC1030-16; lso40	1.7	52	97	46.2	83.5
789	03-FC124	RZM 02-FC124mmaaXA; Sp19	2.8	9	75	15.3	60.8
790	02-FC1015	RZM 02-FC1015mmaaXA; Sp20	2.4	22	85	27.1	72.5
791	03-FC1014-22	Inc. 01-FC1014-22(A,aa); lso4	2.2	37	89	37.2	76.0
792	03-FC123-31	Inc. 01-FC123-31(A,aa); lso78	3.7	9	48	16.3	44.0
793	R378H74	02-1015HO X R178; Sp7	2.2	41	83	38.7	69.6
794	R378H73	02-124HO X R178; Sp7	2.9	23	70	27.7	57.5

[1]Disease Index is based on a scale of 0 (=healthy) to 7 (= plant dead).
[2]Percent of healthy roots (disease classes 0 and 1 combined).
[3]Percent of diseased roots likely to be taken for processing (disease classes 0 through 3 combined).
[4]Percentages were transformed to arcsin-square roots to normalize the data for analyzes.
[5]P=0.05
[6]FC901/C817 - susceptible check
[7]FC705/1 - highly resistant check

**BSDF Project 441 – Cercospora Leaf Spot Research and
Breeding for Cercospora and Curly Top Resistance**
L. Panella & L. E. Hanson
Fort Collins, Colorado

Justification for the Research

This element of the breeding program at Fort Collins is devoted to the development of germplasm with resistance to more than one sugar beet disease and improved agronomic characteristics. It is built on germplasm developed at Fort Collins over the last fifty years for combined resistance to Cercospora leaf spot and the curly top virus. This is an integrated breeding program with greenhouse and laboratory studies, and a field program based on testing in an artificial epiphytotic created in the unique Fort Collins environment. It involves close collaboration with the other USDA-ARS sugar beet programs in the U.S. and sugar beet seed industry customers. The major goals of this program are: 1) the development of sugar beet germplasm with resistance to more than one disease and excellent agronomic characteristics; 2) the improvement of breeding techniques, traditional and molecular, to develop this germplasm; and 3) an increased understanding of the sugar beet/pathogen interactions to improve management practices of these diseases in sugar beet production areas. Genetic information developed during this research will be used to execute additional cycles of pathogen inoculation, plant selection, and recombination among germplasms that we have in our leaf spot improvement program. Results of these tests will be the basis of decisions about specific germplasm, i.e., retain, discard, recombine, release, etc. Germplasms likely to be useful for variety improvement will be identified and released for use by other sugar beet breeders.

Increased resistance to *Cercospora* continues to be an extremely important goal. The need for fungicides would be greatly reduced, if the level of resistance available in most Cercospora-resistant experimental lines were present in commercial hybrids (along with good sugar and seed yield). That continued improvement in genetic resistance to this serious pathogen still is needed, is evident by the occurrence of *Cercospora* strains that are resistant or increasingly tolerant to our most potent fungicides. Additionally, some of these fungicides may be removed from the market because of their perceived or real threat to the environment. In many areas where Cercospora leaf spot is a problem, the curly top virus also causes significant losses. And, there are some growing areas in which combined resistance to Cercospora leaf spot, Rhizomania, curly top, Rhizoctonia root rot, and other diseases are desirable. Germplasm is needed with combined resistance to these diseases, along with good combining ability for yield components.

2004 Field Research on Cercospora Leaf Spot of Sugar Beet

The breeding program in Fort Collins has created an annual artificial epiphytotic through inoculation with *Cercospora beticola* for over forty years. This epiphytotic has been used to evaluate and select for resistance to leaf spot caused by *C. beticola*. We have been pleased to participate in and lead this cooperative research project between the ARS, Colorado State University, and the BSDF.

In 2004 the project primarily involved field studies conducted at the Irrigation Research Center near Yuma, CO. Randomized complete-block designs, with three replications, were used to evaluate commercial and experimental entries. Internal controls included a highly susceptible synthetic (SP351069-0) and a resistant check (FC504CMS/FC502-2//SP6322-0). Two-row plots were 12 feet long, with 22-inch row spacing and an 8- to 10-inch within-row plant spacing. The trial was planted on April 21. Hail and winds hit the plots in early May and caused severe stand losses. Inoculations of remaining plots were performed on July 8 and July 22. The field was sprayed with

Nortron (April 26), Betamix Progress (May 31, June 9, and June 30), Upbeet (May 31 and June 9), and Stinger (June 9) to control weeds. The field was irrigated as necessary. Hail hit the field a total of five times in 2004. Only two of the original seven experimental plots had enough plants to rate. Evaluations were made on August 27, September 3, 10, and 17, with the peak of the epidemic occurring between the second and third ratings.

The moderate temperatures in the summer of 2004 and low moisture (Figure 1 – Project 904) contributed to a moderate leaf spot epidemic, which did not become severe enough to rate until the end of August. Disease severity increased through the first two weeks of September. By the third rating (September 10), means of the resistant and susceptible internal control were 3.0 and 4.3 (scale of 0-10), respectively across the nursery. In 2003 (September 12), these means were 3.5 and 5.8, respectively. Means of contributor lines in 2004 ranged from 2.7 to 5.3.

2004 BSDF Curly Top Nursery at Kimberly, ID

The 2004 curly top nursery began March 1 with 50 cages of leafhoppers. These cages were maintained through the winter and used to increase cages this year. Cages were increased daily. I can make 6-8 cages from one cage putting between 75 - 100 leafhoppers in each cage. All 500 cages were increased by May 6. The nurseries were once again with Kip Wooten (Kimberly site) and Jerry Dickard. The official trials were on the Kimberly site with 694 entries and the complete nursery was at Dickards with 1937 entries. Both fields were in grain in 2003. Both fields were measured and soil sampled the middle of March. Leonard Kerbs, Ag Manager for Amalgamated Sugar, reviewed the soil samples and recommended that 40 lbs of 11 -52 - 0 per acre be applied to both fields. Terry Brown began to receive seed the first part of April. After seed was received, it was completely checked.

Telone was applied to both fields on May 3. Betaseed has built a Telone applicator and Dan Drummond (Betaseed) applied the Telone at 20 gallons per acre. Both fields were disked to seal the soil after the application. Both fields were bedded in the middle of May. After all of the seed was received, planting plans were developed and the seed boxed for planting. The Kimberly field was planted the morning of June 7 and the Dickard field was planted the afternoon of June 7 and finished June 8. As in the past, the University of Idaho's Milton cone planter was used. Each plot was two rows wide and 13 feet long. On the Dickard field the official trials were planted at the beginning of the test and the USDA and seed company material were planted at the end. The official trials were planted in the same order in both fields. Sprinkler pipe was put out on both fields. The Kimberly field was watered for 12 hours on June 11 and 8 hours on June 18. The Dickard field was watered 6 hours on June 10, 14 and 18. Both fields were watered a final time with the sprinkler pipe on June 25 in preparation for thinning and weeding. The fields emerged very well except for the US 41. Monohikari was used in place of US 33 as a susceptible control. Five foot alleyways were cut in both fields June 28-29. The Dickard field was cultivated on June 30. The Kimberly field was thinned and weeded on June 30 - July 2 and the Dickard field was thinned and weeded July 2 - 7. Both fields were cultivated and the water put back on.

The leafhoppers were put on virus-infected plant beginning July 8. Some of these plants were used twice due to a poor survival rate. As last year, a the leafhoppers were moved using the BSDF vacuum pump, to gather the leafhoppers at the greenhouse, and a rented, refrigerated

trailer – this procedure worked very well. The trailer was kept at 50 degrees and stations were set up inside the trailer to catch the leafhoppers. Leafhoppers were released in the Kimberly field on July 13 at 1.5 leafhoppers per plant. Leafhoppers were released in Dickard's field on July 14 in the official trials and company material at 1.5 leafhoppers per plant and on July 15 in USDA material at 1 leafhopper per plant. Leafhoppers were spread by going over the field dragging a trap four times a day for one week. The fields were weeded July 21 - 26 and both fields cultivated July 27. The leafhoppers were sprayed and killed on August 4.

The first rating was taken on August 16. The official trials were rated by Linda Hanson, Stacy Camp, Paul Foote, and Terry Brown. Carl Strausbaugh and Anne Gillan, new USDA scientists observed the rating. John Gallian from the University of Idaho took a sample of curly top from the Kimberly field to find out which strains of curly top were present in the nursery. The preliminary report shows that there is a combination of the Logan and CFH strains of curly top. The second rating was taken on August 30. The official trials were rated by the same people that took the first rating.

On August 30 representatives for Seedex, Betaseed, Stacy Camp, Bob Lewellen, Carl Strausbaugh, Anne Gillan, Tom Schwartz and Terry Brown meet at the Kimberly field to discuss the need for two sites for the official trials. The increase cost of the nursery, and being at capacity for increasing leafhoppers, makes it difficult to maintain two fields. It was decided to continue to plant two tests of official trials but only infect one test.

A third rating was taken on September 13. This rating was taken by Carl Strausbaugh, Stacy Camp, Paul Foote, and Terry Brown. This was a good time to rate because there was more separation than in the first and second ratings.

Cercospora/Curly Top-Resistant Populations with Resistance to Multiple Sugar Beet Diseases and Superior Agronomic Characteristics

Germplasm under Development:

Cercospora Leaf Spot/Curly Top Resistant (LSR/CTR) Breeding Populations Currently Under Development.

FC301 population
1) Cercospora leaf spot and curly top resistant monogerm base population from a polycross of FC607 and FC604 with two Salinas germplasms 2859 and 2890 **(Tested in Salinas as FC123)**.
   a) 2890 (sp) = 0790 *mm aa* x 1890 (Salinas); is seed from *aa* plants open pollinated by A-plants. 0790 = population-790 cycle 5 synthetic by S_1 progeny, *aa*, mm, O-type, good combining ability, adapted to California, S^f. 1890 = BC population to population 790 to get Rz equivalent, remains variable for M-:mm, Rz-:rzrz, etc.
   b) 2859 m (sp) = 1859, 1859R aa x A- (Salinas); Released in 1992 as C859. S^f, similar to 2890, but should have higher curly top resistance. Segregates and variable for M-:mm, Rz-:rzrz, A-:aa, predominant background is lines like C563.

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2) Cercospora leaf spot and curly top resistant multigerm base population from a polycross of FC902 with two Salinas germplasms 278 and 4918.
   a) 278 (Iso 83) = RZM R078; R278 is Rz (segregates Rz--:rzrz) version of C46. It should be S^sS^s, MM.
   b) 4918 (sp) = RZM 3918aa X A-, 142 aa plants; This is an increase of released material C918. It should be Multigerm, over 75% S^f and segregating for A-, R-, Rz-, VY, CT, Erw, & PM.

3) 20021037; (Best FC LSR x Best EL LSR) x CR011 (Salinas LSR/RhzmR) [(20011001 x 2001A031)blk F_2] – tested in Salinas as 04-FC1037
   a) Salinas CR9110 = more broad based rhizomania and leaf spot resistant population, will segregate A- and S^f-
   b) 20011001 = (Best Fort Collins leaf spot resistant x Best East Lansing leaf spot resistant), population cross and bulk made using hypocotyl color

4) 20021038; (Best FC LSR and EL LSR) x CR910 – [(20011001 x 2001A032)blk F_2] – tested in Salinas as 04-FC1038.
   a) Salinas CR910 = fairly inbred rhizomania and leaf spot resistant population, will segregate A- and S^f-
   b) 20011001 = (Best Fort Collins leaf spot resistant x Best East Lansing leaf spot resistant), population cross and bulk made using hypocotyl color.

5) Cercospora leaf spot and curly top resistant multigerm, self-incompatible base population from a polycross of FC607 x [SR87, MonoHy A4, MonoHy T6, & MonoHy T7]

6) Seed from FC709-2 x FC907 was sent to Larry Campbell at Fargo to cross to Sugar beet root maggot resistant germplasm to develop a population that will produce pollinators with resistance to Rhizoctonia, Cercospora, and Root maggot.

Progress in 2004

Advanced breeding lines of *Cercospora* resistant germplasms were lost in the ARS leaf spot nursery at Yuma due to the hail. These lines will be replanted this year. They are part of the resistant germplasm development effort in which a new germplasm should be released from the "pipeline" every two to four years. The above populations currently are in different stages of development.

1) FC301 was released from this population; other selections from ½ sib progeny rows based on combined leaf spot and curly top resistance (FC607&FC604/2859&2890) of the monogerm (FC123mm) and multigerm (FC123MM) population were planted in the 2003 mother root nursery for increase. Material sent to Salinas, CA and showed good rhizomania resistance and progeny families have been selected sucrose. Sib-lines to FC301 that have undergone reselection for resistance to Cercospora leaf spot are being increased for release in 2006 or 2007.

2) Plants (F_2) from the CTR/LSR multigerm cross (2 above – FC902/278/4918) were tested for resistance to Rhizoctonia and Cercospora and recombined. This seed has been bulk increased

and crossed with a number of other leaf spot, rhizomania resistant and high sources populations. The resulting population will be a source of curly top resistant multigerm pollinators with leaf spot and rhizomania resistance. This cross was planted in the Salinas rhizomania resistance nursery for selection and also has been selected for agronomic performance and recombined. It will be tested and evaluated for release or re-selection.

3) Seed of 04-FC1037 was received from R.T. Lewellen in Salinas after selection for sucrose and rhizomania and will be evaluated for Cercospora resistance.

4) Seed of 04-FC1038 was received from R.T. Lewellen in Salinas after selection for sucrose and rhizomania and will be evaluated for Cercospora resistance.

5) Plants (F_2) from the Fort Collins and Fargo joint project (3 above – FC607 x [SR87, MonoHy A4, MonoHy T6, & MonoHy T7]) were grown in the breeding nursery and these roots were planted in Masonville selfed, taking advantage of the 'pseudo self-fertility' that occurs in this environment. This selfed seed was progeny tested in 1999 and the most resistant families were recombined and are being tested and evaluated for release. This population will be a source of highly leaf spot resistant multigerm pollinators with curly top resistance and good combining ability for agronomic traits

6) Seed from (FC709-2 x FC907)F_2 has been sent to Larry Campbell at Fargo to cross to Sugar beet root maggot resistant germplasm and be selected for Cercospora resistance. Sib lines are being reselected this year for Cercospora resistance and this germplasm will be released to provide pollinators with resistance to Cercospora leaf spot and the sugar beet root maggot. Remnant seed from this population will be reselected for Rhizoctonia resistance. The resulting population will provide pollinators with resistance to Rhizoctonia root rot and the sugar beet root maggot.

Table 8. 2004 Curly Top Nursery in Kimberly, ID.				
Source	**Designation**	**Aug 16**	**Aug 30**	**Sep 13**
19931017	Susceptible Check - FC901/C817	3.33	4.00	4.33
1996A008	Beta G6040 - Resistant Check	3.00	3.00	3.33
	Experiment Mean	3.57	4.11	4.59
	LSD $_{0.05}$	0.88	1.39	1.45
	CV	15.4	21.0	19.5
19911042HO	FC402	3.00	3.00	3.00
19911041HO1	FC401CMS	3.00	3.00	3.00
1986A046	FC606 (4x)	3.00	3.00	3.00

Table 8. 2004 Curly Top Nursery in Kimberly, ID.

Source	Designation	Aug 16	Aug 30	Sep 13
19931017	Susceptible Check - FC901/C817	3.33	4.00	4.33
1996A008	Beta G6040 - Resistant Check	3.00	3.00	3.33
	Experiment Mean	3.57	4.11	4.59
	LSD $_{0.05}$	0.88	1.39	1.45
	CV	15.4	21.0	19.5
20001025HO1	FC604CMS	3.00	3.00	3.00
2003A039	FC301 pop - 01-FC123A	3.00	3.00	3.00
19911043HO1	FC403CMS	3.00	3.00	3.00
1986A045	FC606CMS (4x)	3.00	3.33	3.33
20001022	FC710(4X) - LSR Tetraploid	3.00	3.00	3.33
1978A044	FC606	3.00	3.00	3.33
1996A008	**Beta G6040 - Resistant Check**	**3.00**	**3.00**	**3.33**
20021023	FC123mm	3.00	3.33	3.67
20021022	FC123MM	3.67	3.67	3.67
2003A033	FC201	3.33	3.33	3.67
2003A035	FC201 pop - 01-FC1014A	3.33	4.00	3.67
20001025HO	FC604	3.00	3.33	3.67
2002A035	HM9155- high sucrose control	3.00	3.67	3.67
1979A067	FC607	3.33	3.33	3.67
2001A013	EL0204	3.00	3.67	3.67
19911043HO	FC403	3.33	3.33	4.00
20011008HO	FC502-2	3.00	4.00	4.00
20001017	FC720-1	3.33	3.67	4.00
19831028HO	FC506	3.33	3.67	4.00
2003A034	FC301	3.33	4.00	4.00
19941029HO	FC401	3.00	3.67	4.00
19931005HO1	FC721CMS	3.33	3.67	4.00
20011007	F3 LSR MM x RhzcR/LSR (907 x 709-2)	3.33	4.00	4.00
20011024	CTR/LSRmmpop; FC607, FC604, 2890, & 2859	3.33	3.33	4.00
1986A048	FC607 (4x)	3.33	3.33	4.00
19911037	FC719	3.33	3.33	4.00
19741026H	High Sucrose x maritima	3.67	4.33	4.00
19931012	FC901	3.00	3.33	4.00
2003A045	CP08 (Salinas)	3.33	3.67	4.00
19931017	**Susceptible Check - FC901/C817**	**3.33**	**4.00**	**4.33**
19971019	FC716	3.67	4.00	4.33
19911042HO1	FC402CMS	3.33	3.33	4.33
1997A051	FC607CMS	3.33	3.67	4.33
2003A043	CR311 (Salinas)	3.67	4.00	4.33
19931024	FC701	4.00	4.00	4.33
1986A047	FC607CMS (4x)	3.33	4.00	4.33
19951016HO	FC723 – EL44/FC708 mm	3.67	4.00	4.33
19731028HO	FC902	3.00	3.67	4.33
19921019	FC729 – FC712/A4, 3 cycles Rhizoc, MM	3.67	4.00	4.33
19921025	FC728	3.00	3.67	4.33
19951016HO1	FC723 CMS – EL44/FC708 CMS	3.00	3.67	4.33
831085HO	FC708	3.00	4.00	4.33

Table 8. 2004 Curly Top Nursery in Kimberly, ID.

Source	Designation	Aug 16	Aug 30	Sep 13
19931017	Susceptible Check - FC901/C817	3.33	4.00	4.33
1996A008	Beta G6040 - Resistant Check	3.00	3.00	3.33
	Experiment Mean	3.57	4.11	4.59
	LSD $_{0.05}$	0.88	1.39	1.45
	CV	15.4	21.0	19.5
19771067HO	FC504	3.50	4.00	4.50
19921008	FC725	3.33	3.00	4.67
19971020	FC907-1 – FC607/FC701 BC4 - 1 cycle of RhzcR sel	3.67	4.00	4.67
19941024	FC710	3.33	4.00	4.67
20001016H2	(FC708CMS X FC709-2)	3.67	4.33	4.67
19961010HO	FC722-1 – C718/FC708	3.33	3.33	4.67
19881032H	FC712	3.67	3.67	4.67
2004A008	EL51	4.00	4.33	4.67
20031014	(SucroseMM x PI540605)F3	3.67	4.00	4.67
20011002bbMS	LSR (France) x SucroseMM - aa biennial segregants	3.33	4.33	4.67
19931005HO	FC721	4.00	4.00	4.67
20011001	LSR Polycross with East Lansing material	3.33	4.00	4.67
20021018HO1	FC712/Monohy A4 CMS equivalent	3.33	4.00	4.67
20031013	SucroseMM x PI35826 LSR Fodder Beet	3.67	4.33	4.67
19751080H	FC703	3.67	4.00	5.00
20021018HO	FC712/Monohy A4	3.67	4.00	5.00
19911026HO	FC715	4.00	5.00	5.00
19921021	FC703-5	4.00	5.00	5.00
19801059H	FC701-6	4.00	4.67	5.00
19821087	FC711	4.00	4.67	5.00
20001018	FC704	4.00	4.00	5.00
19991016	FC702/2	3.67	4.00	5.00
19991015	FC801	3.67	4.67	5.00
20001019	FC705	3.67	4.33	5.00
1987A026	AD1 (4x)	3.67	4.00	5.00
19991017	FC703	3.67	4.33	5.00
19811055H	FC702-6	4.00	4.67	5.33
20001020	FC706	3.67	4.00	5.33
2001A016	SR94; WC960448	4.33	5.00	5.33
2001A020	94HS25; WC960452; smooth root	4.33	5.00	5.33
19891037	AD2 (4x)	4.00	4.67	5.33
19971018	FC712(4X)	3.67	4.67	5.33
20011002bbPF	LSR (France) x SucroseMM - A_ biennial segregants	4.00	4.33	5.33
20011009	FC702-4(4X)	3.33	4.33	5.33
2001A015	SR95; WC970308	3.67	4.67	5.33
19921022	FC702-7	4.00	4.67	5.33
19821051H2	**LSR**	3.67	4.67	5.33
2001A014	SR96; WC980437	4.00	4.67	5.33
19951017	FC727	3.67	4.00	5.67
20001007	LSR w/ Fargo	4.00	5.33	5.67
19961014	FC724 New Release	4.00	4.33	5.67
19991018	FC709	4.33	5.00	5.67

Table 8. 2004 Curly Top Nursery in Kimberly, ID.

Source	Designation	Aug 16	Aug 30	Sep 13
19931017	Susceptible Check - FC901/C817	3.33	4.00	4.33
1996A008	Beta G6040 - Resistant Check	3.00	3.00	3.33
	Experiment Mean	3.57	4.11	4.59
	LSD $_{0.05}$	0.88	1.39	1.45
	CV	15.4	21.0	19.5
19961010HO1	FC722 CMS – C718/FC708 CMS	4.00	5.00	5.67
19951029	AD3 (4x)	3.67	5.33	5.67
19911032	FC718	4.00	4.67	5.67
20001021	FC707	4.33	5.33	5.67
19931010	FC726	4.00	4.67	6.00
20001016H	FC709-2	4.67	6.00	6.00
19981025	FC717	3.67	5.67	6.00
19831083	FC705/1	4.00	4.50	6.00
19761068H	FC701-4	4.33	4.67	6.00
19751132	Russian Multi-germ Germplasm Pool	4.33	5.67	6.33
2002A037	Beta 6045	4.00	5.33	6.67
2002A036	Monohikarí - high sucrose control	4.67	6.00	6.67
19751099H	L-19	3.67	5.67	7.00

Table 9. Experiment 4A, 2004. Leaf Spot Evaluation of USDA-ARS Salinas contributed lines.

Entry	Identification	Disease Index[1]			
		August 27th	September 3rd	September 10th	September 17th
	LSD$_{0.05}$	1.20	1.35	1.24	0.97
358	LSS [2] (931002)	4.0	4.3	4.7	4.0
359	LSR [3] (821051H2)	1.0	2.7	3.0	2.0
	Trial Mean	2.7	3.4	3.7	3.1
321	Beta 4430R	5.7	5.7	5.3	5.0
322	Monohikari	2.0	3.0	3.3	3.7
323	03-SP 22-0	2.3	3.0	3.0	2.7
324	Y390	3.3	3.7	4.0	3.3
325	Y391	3.3	5.0	4.7	3.7
326	Y392	3.7	4.2	4.3	3.7
327	Y393	3.3	3.3	3.7	3.3
328	Y375	2.7	4.0	3.7	3.7
329	R321	3.3	3.7	4.0	3.7
330	CR311	2.0	2.7	3.0	2.3
331	3941	3.7	4.3	4.3	3.3
332	3933	3.2	3.8	3.7	2.7
333	3933-107	2.7	3.7	3.7	3.0
334	3933-113	2.7	2.7	3.3	3.0
335	3933-118	3.5	4.5	5.0	3.5
336	2933-14	1.3	2.3	3.0	2.3
337	03-FC1033-15	2.7	3.3	4.0	3.3

Entry	Identification	Disease Index[1] August 27th	September 3rd	September 10th	September 17th
	LSD$_{0.05}$	1.20	1.35	1.24	0.97
358	LSS [2] (931002)	4.0	4.3	4.7	4.0
359	LSR [3] (821051H2)	1.0	2.7	3.0	2.0
	Trial Mean	2.7	3.4	3.7	3.1
338	03-FC1033-16	2.3	3.7	4.3	3.7
339	CR311-6	1.3	2.3	2.7	2.3
340	CR311-41	2.5	2.5	3.0	2.0
341	CR311-88	2.7	3.3	3.0	2.3
342	CR310-14-2	2.5	2.5	3.5	3.0
343	03-FC124	3.3	3.3	4.0	3.0
344	02-FC1015	3.3	3.7	4.3	3.3
345	03-FC1014-22	2.0	2.7	3.7	2.7
346	03-FC123-31	3.3	3.7	4.0	3.3
347	R378 H74	2.7	3.7	3.7	3.3
348	R378 H73	2.3	2.7	3.0	3.0

Table 9. Experiment 4A, 2004. Leaf Spot Evaluation of USDA-ARS Salinas contributed lines.

[1]Disease Index is based on a scale of 0 (=healthy) to10 (=dead).
[2]The Leafspot Susceptible Check is SP351069-0.
[3]The Leafspot Resistant Check is ((FC504CMS x FC502/2) x SP6322-0).
 Means and LSD values were calculated with missing data. Lines 335, 340, and 342 were two reps rather than three.
 LSD and experimental mean values also were calculated with an additional nine entries

BSDF Project 443 – Pre-breeding: the Introgression of New Sources of Cercospora Leaf Spot Resistance from *Beta Vulgaris* ssp. *maritima* and other Exotic Sources into Sugar Beet-type Populations.

Lee Panella
Fort Collins, Colorado

A major emphasis of the research mission of the USDA-ARS plant scientists is the collection, documentation, characterization, evaluation, regeneration (maintenance), distribution, and utilization of plant germplasm, especially Plant Introduction (PI) accessions in the USDA-ARS National Plant Germplasm System (NPGS). The Sugar Beet Research Unit at Fort Collins is coordinating the national program for *Beta* germplasm evaluation. In addition to the evaluation for Rhizoctonia and Cercospora resistance, it is crucial that the ARS scientist be involved in the long rang, high risk research problems involved in sugar beet 'germplasm enhancement' or 'pre-breeding' from exotic germplasm or wild relatives. This is an important component in the overall sugar beet improvement effort of the Fort Collins Sugar Beet Research Unit.

Justification for Research:

Cercospora leaf spot (caused by the fungus *Cercospora beticola* Sacc.) is one of the most widespread diseases of sugar beet and is a serious problem in many sugar beet production areas throughout the U.S. The disease damages the leaves, which, consequently, reduces root yield, percent sucrose of roots, and purity of the extracted juice. Cercospora leaf spot currently is controlled by combining spraying with commercial fungicides and the use of disease tolerant germplasm. The development of Cercospora leaf spot resistant sugar beet lines and hybrids with greater levels of host-plant resistance offers a more sustainable solution to this disease problem.

If the level of resistance available in some Cercospora-resistant experimental breeding lines were present in commercial hybrids **(along with good sugar and seed yield)**, the need for fungicides could be greatly reduced. That continued improvement in genetic resistance to this serious pathogen is still needed is evident by the occurrence of *Cercospora* strains that are tolerant to our most potent fungicides. Additionally, some fungicides may be removed from the market because of their perceived or real threat to the environment.

Finally, the genepool for resistance to Cercospora leaf spot is extremely narrow. Many of the resistant lines are highly inbred, therefore, closely related to one another, and stem from germplasm coming out of Italy in the early 1900s. In the germplasm developed at Fort Collins, continued inbreeding has increased the level of disease resistance, but at the cost of plant vigor. Over the long term, a secure, sustainable response to this disease requires commercial quality hybrids with good host-plant resistance.

Summary of Literature Review:

Cercospora leaf spot (CLS) has been an intermittent problem in sugar beet growing areas of the United States where the summers can be hot and humid (Red River Valley, Michigan, Ohio, and, less often, Great Plains growing areas and California). It has been estimated that a severe epidemic can cause up to a 42% loss of gross sugar (Smith and Martin, 1978; Smith and Ruppel, 1973), or up to a 43% relative dollar loss (Shane and Teng, 1992).

Resistance to CLS has long been a goal of the USDA-ARS sugar beet research program at Fort Collins and researchers there developed the techniques necessary to manage the screening nurseries in such a way as to promote the development of the disease (Ruppel and Gaskill, 1971). A careful crop rotation (sugar beet-barley-barley-barley-sugar beet) and the arid climate and low relative humidity have allowed this to be done in such a manner that there are rarely high enough levels of any other disease present in the leaf spot nursery to confound the results. The resistance to CLS could more accurately be described as a tolerance, rather than true resistance. Tolerance or "field resistance" means that, although some symptoms of the disease are present, the plant still is able to perform well (Fehr, 1987 p.307).

Much of the Cercospora-resistant germplasm in use today came out of Munerati's program in Italy, in which B. vulgaris spp. maritima was the source of resistance genes (Lewellen, 1992). In this genetic source, there are an estimated 4 or 5 genes responsible for CLS resistance (Smith and Gaskill, 1970) and broad-sense heritability estimates ranged from 12 to 71% (Bilgen et al., 1969). Narrow-sense heritability estimates of about 24% compared well with realized heritability values, and 44 to 62% of the variation was due environment in this test (Smith and Ruppel, 1974). The large environmental variation has made it difficult to make progress in developing resistance through mass selection. Incorporation of high levels of leaf spot resistance into varieties with superior agronomic performance also is difficult (Smith and Campbell, 1996) and, therefore, commercial resistant varieties require some fungicide application to provide adequate levels of protection against Cercospora (Miller et al., 1994).

A major problem in the development of CLS-resistant sugar beet is the loss of vigor due to the continual inbreeding. Coons (1955) noted this and it has been a concern ever since (McFarlane, 1971). The use of hybrid varieties has ameliorated this problem to some extent, but seed production on the highly inbred O-type males and CMS females still is a problem. This creates an urgent need to continue to develop a broader genetic base in our CLS-resistant germplasm than we have today. Also as commercial hybrid parents become more inbred, the germplasm base from which these inbred parents are developed must have the diversity necessary to provide for maximum gain through heterosis. In addition to broadening the genetic base of the commercial sugar beet germplasm, novel genes for resistance to CLS might lead to transgression of the currently available tolerance to CLS. Simply defined, transgression is when a population contains individuals with a phenotype that is beyond the phenotype found in the parents of the population (de Vicente & Tanksley, 1993).

The USDA-ARS National Plant Germplasm System Beta collection has over 2,000 Plant Introduction (PI) accessions. The germplasm used most often in sugar beet breeding is from Beta vulgaris spp. vulgaris, which includes all of the biennial sugar beet types, or from Beta vulgaris spp. maritima, which contains the closely related wild sea beet and has both annual and biennial types. Germplasm with a biennial flowering habit is easier both to introgress and screen. Beta vulgaris spp. maritima has, nonetheless, been used as a source of resistant germplasm. Much of the CLS-resistant germplasm in use today, which came out of Munerati's program in Italy, had B. vulgaris spp. maritima as the source of resistance genes (Lewellen, 1992). There have been very few new efforts to locate and incorporate other sources of resistance to Cercospora into this narrow germplasm base. Munerati's success, and the research of others, has shown that it can be done if we have the persistence to do it (Bilgen et al., 1969; Doney, 1993; Lewellen, 1995).

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Objectives:

1. The formation of long range breeding populations through the introgression of Cercospora resistant germplasm from "exotic" sources (Beta vulgaris ssp. maritima, fodder beet, foreign sugar beet landraces from the PI collection, etc.).

2. The development of germplasm populations from these long range populations that are of sufficient agronomic quality to be of use to commercial breeders. They will be a source of leaf spot resistance with different genetic backgrounds.

3. The development of techniques (both traditional and molecular) to more efficiently introgress the exotic germplasm into sugar beet breeding populations.

Materials and Methods:

Artificial field inoculation with Cercospora beticola and leaf spot scoring will be used to identify the resistant germplasm sources and make selections in the developing populations. The exotic materials will be crossed into sugar beet populations that have been selected for agronomic quality (recoverable sucrose yield). These sucrose populations are based on old commercial varieties – i.e., MonoHy T6, A7, A4 and breeding lines from American Crystal Sugar Co. and Seedex, Inc. – and USDA-ARS developed germplasm such as L-19 (WC9127OM) and East Lansing smooth root germplasm , SR87. Other parents include high sucrose germplasm from Poland and other Eastern European countries. Salinas parent '3859' was used to produce populations that are self-fertile (S^f) and segregating for nuclear male sterility (A-:aa).

Hybrid populations will be handled in the following manner: 1) Following the initial cross, a population will be random mated (using aa females because of the self-fertility) for three to four generations to break up linkage groups and remove annual plants. 2) Sugar beet-type mother roots will be selected, selfed, and progeny tested for agronomic performance and disease resistance. 3) Selected roots will be recombined (and backcrossed if desirable) and re-selected until they ready for release. Molecular markers (RFLPs, RAPDs, SSRs, AFLPs, etc.) as they become available will be used to expedite the backcrossing program and to follow the change in allele frequencies in the selected populations. Advanced populations will be released to the sugar beet seed industry.

Time Line of Anticipated Accomplishments:

Development of a resistant germplasm line generally takes seven years. A longer time will be necessary to incorporate disease resistance from more exotic sources. Because this is a new program, it will take time for the first germplasm to make it through the process. Once that happens, there will be a "pipeline" of germplasm in various stages of development and the release of new germplasm will occur every two to four years. The incorporation of exotic sources into agronomically acceptable germplasm is a long term proposition - results will not appear overnight. This is the type of long-term germplasm development that ARS is well suited to perform.

Research Progress 2004

We are working with the eighteen populations listed below (Table 3) and have increased or made crosses in these populations. All of the male parents are germplasm that have been identified as having resistance to *Cercospora beticola* (causal agent of Cercospora leaf spot). The female parents are from a population developed to have high sucrose yield potential. These sucrose populations are based on old commercial varieties – i.e., MonoHy T6, A7, A4 and breeding lines from American Crystal Sugar Co. and Seedex, Inc. – and USDA-ARS developed germplasm such as L-19 (WC9127OM) and East Lansing smooth root germplasm , SR87. Other parents include high sucrose germplasm from Poland and other Eastern European countries. Salinas parent '3859' was used to produce populations that are self-fertile (S^f) and segregating for nuclear male sterility (*A-:aa*). The families from various crosses are in different stages of development and evaluation. At the F_3 stage, when sufficient seed is available, we are beginning field screening and selection. Seed of these families has been bulk increased and is beginning to be evaluated. All of the early generation populations show some annual plants in our environment.

We are re-crossing some of those from which we obtained insufficient F_1 seed. Plants from those populations producing some biennial plants are being vernalized for 90 days and the populations are being increased (i.e., random mated using the genetic male sterility where possible). The annuals will be handled in a similar fashion once the F_1 populations have been increased. All will be cycled through at least three cycles of random mating.

The most advanced populations were planted in the 2004 mother root nursery for increase and to select non-bolting types. They were also planted in the leaf spot nursery at Yuma. CO. This nursery was hailed on four times in 2004 and the trial was lost. They will be screened next summer for resistance to Cercospora leaf spot and curly top. In 2002 (below), Leaf spot evaluations showed good levels of resistance for some of the populations and some also showed resistance to the curly top virus. All of the early populations are still segregating for biennial growth habit, easy bolting, and other wild traits.

Table 10. List of germplasm used in developing Cercospora leaf spot resistant populations and the stage of each of the populations. Those populations highlighted in yellow have been increased in 2004 and those in green will be increased in 2005.

♀ parent	Donor (♂) Designation	Name or Origin (♂)	% Bolting (♂) no induction 1996 FC, CO	F₁ Population	F₂ Population	F₃ Population	F₄ Population	F₅ Population	F₆ Population
961005	PI 535826	Giant Poly	20%	971021H2	981031	991026	20011027MS	20031013	20051002
961005 19991024H2	PI 535833	Saturn	0%	--- 20031001H2					
961005	PI 540593	WB 847	0%	971023H2	20021026	20051001			
961005	PI 540596	WB 850	70%	971024H2	981032	20011002bbPF 20011002bbMS 20011002B-	20051004		
961005	PI 540605	WB 859	25%	971025H2	20011054	20031014	20051005		
961005	PI 535843	PN MONO 1	100%	971026H2[1]					
961005	PI 540575	WB 829	100%	971027H2[2]					
961005	PI 540599	WB 853	50%	971028H2	981033	20011045bbPF 20011045bbMS	20051003		
961005	BGRC #32375 (*B. v. maritima*)	Greece	annual	971029H2	20011036				
961005	BGRC #36538 (*B. v. maritima*)	Greece	annual	971030H2[3]	20011037				
851046HO	BGRC #45511 (*B. v. maritima*)	Greece	annual	981001H3	20011038B_ 20011038bb				
961005 19991024H2	BGRC #45511 (*B. v. maritima*)	Greece	annual	--- 2001046H2	20021036B_ 20021036bb	20051006			
851046HO	*BGRC #45516* (*B. v. maritima*)	Greece	annual	981002H3	20011039B_ 20011039bbPF 20011039bbMS				

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Table 10. List of germplasm used in developing Cercospora leaf spot resistant populations and the stage of each of the populations. Those populations highlighted in yellow have been increased in 2004 and those in green will be increased in 2005.

♀ parent	Donor (♂) Designation	Name or Origin (♂)	% Bolting (♂) no induction 1996 FC, CO	F$_1$ Population	F$_2$ Population	F$_3$ Population	F$_4$ Population	F$_5$ Population	F$_6$ Population
961005 19991024H2	*BGRC* #45516 (*B. v. maritima*)	Greece	annual	-- 20021033H2					
961005	BGRC #48810 (*B. v. maritima*)	Tunisia	annual	19981003H2	20011040B_ 20011040bb	20021030B_ 20021030bb	20031038bb		
851046HO	BGRC #48810 (*B. v. maritima*)	Tunisia	annual	981003H3	200110141B_ 200110141bb				
961005	BGRC #48819 (*B. v. maritima*)	Tunisia	annual	981004H2	20011042B_ 20011042bb	20021031B_ 20021031bb	20031039B_ 20031039bb		
961005 19991024H2	BGRC #48819 (*B. v. maritima*)	Tunisia	annual	-- 20021034H2					
961005 19991024H2	BGRC #51430 (*B. v. maritima*)	Greece	annual	-- 20021035H2					

[1] Only 16 seed balls produced.
[2] Only 10 seed balls produced.
[3] Only 60 seed balls produced.
Boxed = into GH/steck field in 2005/06
2005xxxx out of 2004 steck field (2005 seed production number)

SUGARBEET RESEARCH

2004 Report

SECTION C

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CONTENTS

PUBLICATIONS
Abstract of Papers Presented or Published

Klotz, K.L., and Finger, F.L. 2004. Respiration rate of postharvest sugarbeet root is unrelated to respiratory capacity or adenylates. 5th International Postharvest Symposium. Abstract No. S9-43, p. 92.

Respiration is the primary cause of sucrose loss during postharvest storage of sugarbeet (Beta vulgaris L). The factors that regulate respiration in sugarbeet root, however, are largely unknown, although substrate availability, respiratory capacity and adenylate levels have been shown to control respiratory flux in other plant species. The relationship of respiration rate to respiratory capacity and adenylates was investigated in harvested sugarbeet roots stored at 1o and 10o C with or without wounding. Respiration increased in wounded roots stored at 10o C and in wounded and unwounded roots after prolonged storage at 1o C. Differences in total respiratory capacity, cytochrome c oxidase capacity, alternative oxidase capacity, ATP concentration, ADP concentration, ATP:ADP ratio, and phosphorylation potential were also observed between treatments, although changes in these parameters were unrelated to root respiration rate. Additionally, uncoupling respiration from oxidative phosphorylation by treating tissue sections with carbonyl cyanide-m-chlorophenyl hydrazone (CCCP) resulted in no increase in respiration rate. These results indicate that postharvest sugarbeet root respiration is not significantly regulated by respiratory capacity or adenylate levels.

Klotz, K.L., Finger, F.L. 2004. Impact of temperature, length of storage and postharvest disease on sucrose catabolism in sugarbeet. Postharvest Biology and Technology. 43:1-9.

Sucrose catabolism during postharvest storage of sugarbeet (Beta vulgaris L.) root has been the subject of several studies, yet no consensus exists about the contribution of individual sucrolytic activities to postharvest sucrose loss. Because differences in storage temperature, length of storage, and the presence of storage pathogens may have contributed to the discrepant results from earlier studies, the impact of these three factors on sugarbeet root postharvest sucrose catabolism was determined. Sucrolytic activities and soluble carbohydrate concentrations were measured in roots exhibiting no pathological symptoms during storage at 6, 12 and 21o C, and in roots exhibiting severe rotting symptoms due to infection by Penicillium spp. and Botrytis cinerea at 6o C. Sucrose synthase was the predominant sucrolytic activity throughout storage, regardless of storage temperature, length of storage, or pathogenesis, and accounted for more than 90% of the total soluble sucrolytic activity present in roots. In disease free roots, no significant change in sucrose synthase activity, soluble acid invertase activity, insoluble acid invertase activity, glucose concentration or fructose concentration occurred in roots stored at 6 or 12o C, although an increase in sucrose synthase activity and fructose concentration was

observed in roots stored at 21o C. Alkaline invertase activity was impacted by the length of storage and exhibited a transient decline in activity at all storage temperatures. In roots with severe rot, insoluble acid invertase activity declined, sucrose synthase and alkaline invertase activities were unchanged, and soluble acid invertase increased seven-fold. The increase in soluble acid invertase activity was primarily due to the presence of fungal acid invertase isoforms. These results indicate that sugarbeet sucrolytic activities change little during storage, regardless of storage temperature, length of storage, and pathogenesis, and suggest that sucrose synthase, as the predominant sucrolytic activity in stored roots, is central to postharvest sucrose catabolism in sugarbeet roots.

Klotz, K.L. 2005. Anatomy and physiology. In: Biancardi, E., Campbell, L.G., Skaracis, G.N., and De Biaggi, M. Genetics and Breeding of Sugar Beet. Enfield, New Hampshire, Science Publishers, Inc. p. 9-18.

The anatomy of the major organs of sugarbeet and the physiology of the major events in the two-year life cycle of sugarbeet are reviewed. Gross morphology of the root and shoot of sugarbeet during vegetative and reproductive growth is described. Root, leaf, flower and seed anatomy and development are also described. The physiological events of seed germination, the transition from vegetative to reproductive growth, flowering, fertilization, and self-incompatibility are reviewed, and the environmental factors that affect these events are discussed.

Klotz, K.L., and Campbell, L.G. 2004. Sucrose catabolism in developing roots of three beta vulgaris genotypes with differentyield and sucrose accumulating capacities. Journal of Sugarbeet Research. 41(3): 73-88.

The functions of the major sucrolytic enzymes in sugarbeet (Beta vulgaris L.) roots are poorly understood, although a positive association between sucrose synthase activity and root size, and a negative association between soluble acid invertase activity and sucrose concentration have been documented. To test the veracity of these relationships and determine whether any major sucrolytic activities were associated with root yield or sucrose accumulation, the activities of the major sucrolytic enzymes, fresh and dry mass, and the content of sucrose, glucose, fructose and cell wall materials were measured in roots of three B. vulgaris genotypes with differing yield and sucrose accumulating capacities at five stages in their development. Across all genotypes and developmental stages, sucrose synthase activity was positively associated with root mass, water content, and accumulation of cell wall materials. No meaningful association was observed between alkaline invertase, soluble acid invertase and insoluble acid invertase activities and any of the physical or chemical properties examined. Although no new insights into the functions of acid and alkaline invertases were gained, these results indicate that sucrose synthase activity is a reliable indicator of root yield and may be a factor in root sink strength and the promotion of cell wall biosynthesis.

Haagenson, D.M., Klotz, K.L. 2004. **Raffinose synthase is influenced by postharvest storage temperature and duration. Abstracts from ADA, CSSA, and SSSA International Annual Meetings, Oct 31-Nov 4, 2004, Seattle, WA. [CD-ROM]**

Raffinose is an important impurity in sugarbeet (Beta vulgaris L.) processing as it decreases extractable sucrose, and raffinose concentrations may increase with prolonged periods of cold (less than 4°C) during sugarbeet growth and storage. Our objective was to gain a physiological understanding of environmental and biochemical factors controlling sugarbeet raffinose accumulation. Beets were harvested three times (8 September, 23 September, and 29 October 2004), and were stored for 2, 10, or 18 wk at 2 or 6°C, respectively. Root and crown tissues were analyzed for raffinose content, and raffinose synthase transcript abundance and enzyme activities. As expected, sugarbeet root raffinose concentrations increased during storage and were three-fold higher in beets stored at 2°C when compared to 6°C. Raffinose synthase transcript abundance and enzyme activities were highest in roots stored for 2 wk at 2°C. When comparing the three harvest dates, beets harvested 8 September had the highest transcript abundance and enzyme activity after 2 wk of storage at 2°C. Transcript abundance and enzyme activity declined at 10 and 18 wk of storage among all harvest dates, but roots stored at 2°C maintained a 6-fold higher enzyme activity when compared to roots stored at 6°C.

Weiland, J.J. and Koch. G. 2004. **Sugar-beet leaf spot disease (*Cercospora beticola* Sacc.). Molecular Plant Pathology. 5:157-166.**

Leaf spot disease caused by *Cercospora beticola* Sacc. is the most destructive foliar pathogen of sugarbeet worldwide. In addition to reducing yield and quality of sugarbeet, the control of leaf spot disease by extensive fungicide application incurs added costs to producers and repeatedly has selected for fungicide-tolerant *C. beticola* strains. The genetics and biochemistry of virulence have been examined less for *C. beticola* as compared with the related fungi *C. nicotianae*, *C. kikuchii* and *C. zeae-maydis*, fungi to which the physiology of *C. beticola* is often compared. *C. beticola* populations generally are not characterized as having race structure, although a case of race-specific resistance in sugarbeet to *C. beticola* has been reported. Resistance currently implemented in the field is quantitatively inherited and exhibits low to medium heritability.

McGrath, J.M., Shaw, R.S., De los Reyes, B., and Weiland, J. 2004. **Construction of a sugarbeet BAC library from a hybrid with diverse traits. Plant Molecular Biology Reporter. 22:23-28.**

A bacterial artificial chromosome (BAC) library of the 750-Mbp sugar beet genome represented in hybrid US H20 was constructed from *Hin*d III-digested DNA, with an average insert size of 120 kbp. US H20 is a variety grown in the eastern United States. It exhibits heterosis for emergence and yield, presumably because of its

hybridity between eastern and western US germplasm sources. Filter arrays were used to assess the abundance and distribution of particular nucleotide sequences. An rRNA gene probe found that 1.2% of the library carried sequences similar to these highly repetitive and conserved sequences. A simple sequence repeat element $(CA)_8$ thought to be predominantly distributed throughout centromere regions of all chromosomes was present in 1.7% of clones. For more than half of the 28 randomly chosen expressed sequence tags (ESTs) used as probes, a higher-than-expected number of single-copy hybridization signals was observed. Assuming 6× genome coverage, this suggests that many duplicate genes exist in the beet genome.

POLYMERASE CHAIN REACTION (PCR)-BASED DETECTION OF FUNGAL PATHOGENS USING ACTIN GENE SEQUENCES.
Project 620

John J. Weiland

The polymerase chain reaction (PCR) is a DNA based technique for amplifying specific sequences from the genomes of organisms. PCR technology has impacted many fields of biology, including the area of disease diagnosis in both plants and animals. Diagnostics using the PCR are sensitive and highly discriminatory, since they target genome regions whose DNA sequences have diverged throughout evolution. PCR-based diagnostics also require little time for a result to be secured (within one to two days), making them attractive to high-throughput diagnostic laboratories. More recently, exquisite quantitation of pathogens has been made a reality by the added technology of "real-time" PCR, a technology currently being used in our laboratory for the quantitation of gene expression and fungal genomes.

The interests in our laboratory include the development of novel diagnostic tools for disease-causing fungi in sugarbeet with a special emphasis on the highly destructive pathogen *Aphanomyces cochlioides*.. For this reason, we designed our PCR assay for the discrimination of sugarbeet fungal pathogens upon DNA sequences of the actin, ribosomal RNA (rRNA), and mitochondrial cytochrome b genes. The chosen genes harbor sequences that permit that organism to be "fingerprinted" according to that gene sequence. This fingerprinting analysis was applied to Aphanomyces populations that were collected in the U.S. ranging from the northern Red River Valley to (now abandoned) sugarbeet growing regions of Texas. The analysis revealed that *Aphanomyces cochlioides* populations in the central states of the U.S. are genetically uniform. Because of this, we sought to examine *A. cochlioides* isolates from a more localized region that nevertheless has some unique attributes.

Sugarbeet grown in the Southern Minnesota Beet Sugar Cooperative region is, in some cases, rotated with fields of green pea and with table beet. *A. euteiches* is a well known pathogen of peas in this area. In addition, it is known that *A. cochlioides* can infect table beet. We therefore collected soil samples from these regions in 2003 and have produced DNA preparations from 85 single-zoospore isolates from these samples. The genetic diversity of these isolates will be determined and compared to results obtained from previous studies.

In addition to genetic fingerprinting that can be done with PCR technologies, real-time quantitative PCR (qPCR) can be used to quantitate levels of pathogen in infected tissue or in soils. To this end, sequences in the actin genes of *A. cochlioides* and *A. euteiches* were aligned in order to define regions that distinguish the two pathogens. Primer/probe combinations were produced capable of amplifying *A. cochlioides*, but not A. euteiches. The ability for this primer/probe combination to amplify *A. cochlioides* DNA in a quantitative manner is shown in Figure 1. We have just completed DNA extraction from soils known to contain high versus low levels of *A. cochlioides*. In addition, we currently are preparing DNA from tolerant versus susceptible sugarbeet hybrids. Both samples will be used to test the qPCR technique for its ability to quantitate *A. cochlioides* levels.
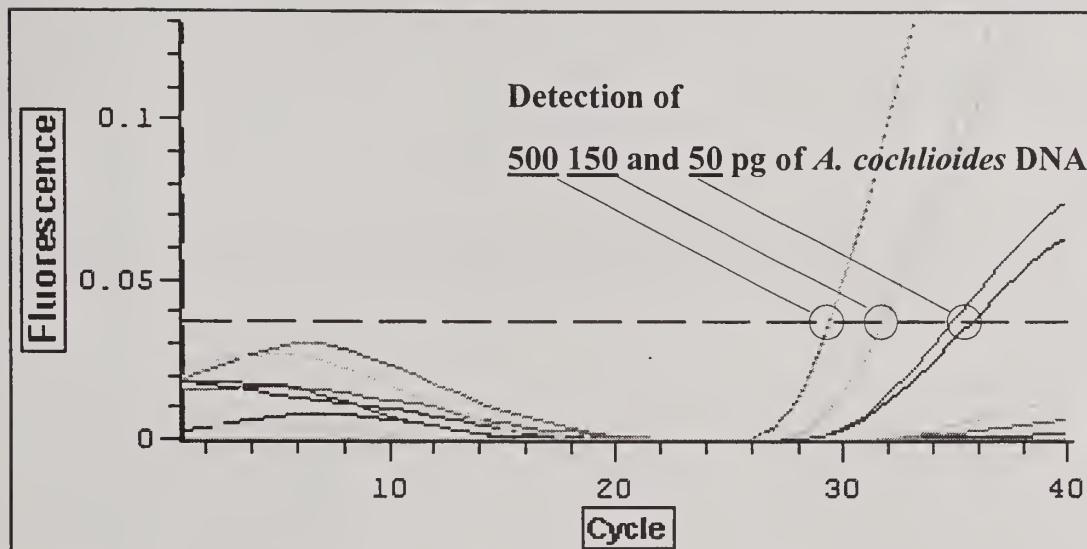
Figure 1   Quantitative detection of *A. cochlioides* using qPCR. Primers specific for actin gene sequences of *A. cochlioides* were coupled with a 6-FAM-labeled probe (Integrated DNA Technologies, Coralville, IA), itself designed for specificity to the pathogen. Dilutions of genomic DNA prepared from *A. cochlioides* strain 19-1z (provided by C.Windels) were targets for the detection process. Digestion of fluorescent probe (100 nM in the reaction) was monitored during amplification in an MJR Opticon 2 (Bio-Rad Laboratories Inc., Hercules, CA) thermocycler.

MECHANISMS OF RESISTANCE IN SUGARBEET TO FUNGAL AND BACTERIAL PATHOGENS
Project 621

John J. Weiland

Enzymes and enzyme inhibitors that accumulate in sugarbeet that is under pathogen stress often are associated with resisting pathogen invasion. Some of these activities are produced to strengthen natural barriers in the plant to pathogen invasion. Others are produces as an arsenal of compounds toxic to the pathogen or as inhibitors of phytotoxins produced by the pathogen. Identification of sugarbeet enzymes, and their corresponding genes, produced in defense against pathogens can further our understanding of the basis for disease resistance. Such knowledge can be used in the selection of germplasm with enhanced pathogen resistance. In addition, the cloning of the genes for defense-related enzymes and inhibitors can lead toward the production of genetically modified (engineered) germplasm for use in sugarbeet breeding programs.

Protease activity secreted in to the culture media by *A. cochlioides* is being investigated as a virulence component in the production of disease in sugarbeet. Proteases are produced in abundance by Aphanomyces species, including those that infect fish and crayfish. Previously in our lab, it was shown that a proteinase inhibitor from lima bean effectively inhibits a subset of the proteases that are seperable using gel electrophoresis. In 2004, we began purifying a ~70 kilodalton protease expressed by *A. cochlioides* that is abundantly produced in infected sugarbeet seedlings. Antiserum is currently being raised against this protease.
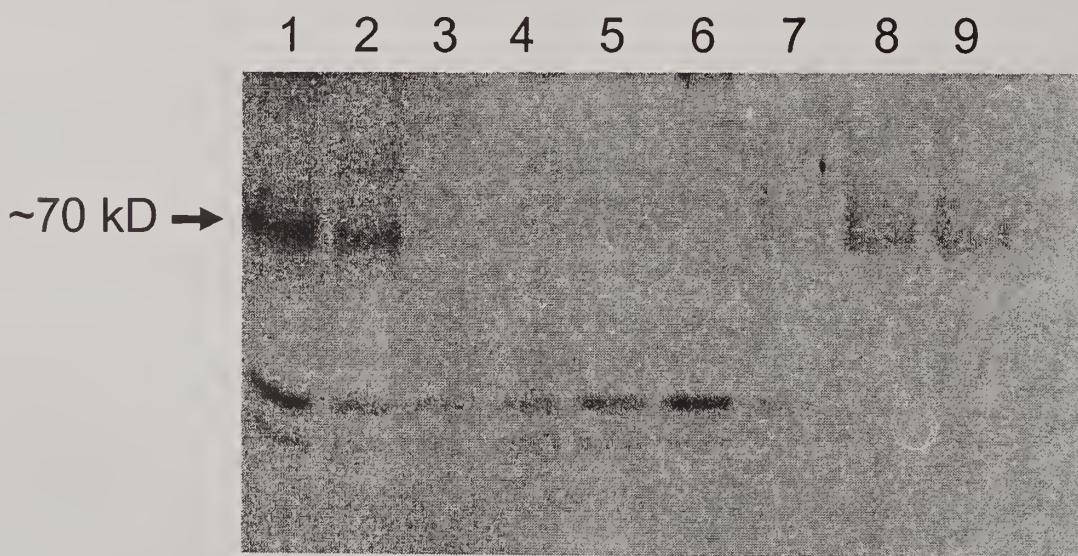
Figure 2 Protease activity secreted by *A. cochlioides*: Purification process. Lane 1, total secreted protease; lane 2 total protease adjusted with chromatography buffer; lanes 3-9, fractions collected from DEAE Sephacel column. Note separation of 70kD protease from the faster-migrating trypsin-like proteases.

Also in 2004, mutants of *Cercospora beticola* that lack cercosporin production were tested in greenhouse inoculations on the sugarbeet variety ACH 9369. These mutants were generated by disruption of a polyketide synthase gene (*ctb*1) already known to be required for cercosporin biosynthesis in *C. kikuchii* and *C. nicotianae*. These mutants produce only background levels of cercosporin in culture relative to the wild-type isolate Cb303B (Figure 3). The mutants still produce leaf spot lesions, but at a lower rate of spot density and with a lower degree of lesion expansion. The experiments convincingly show that cercosporin production is important for disease on sugarbeet, but that other factors additionally are involved in causing leaf spot disease.
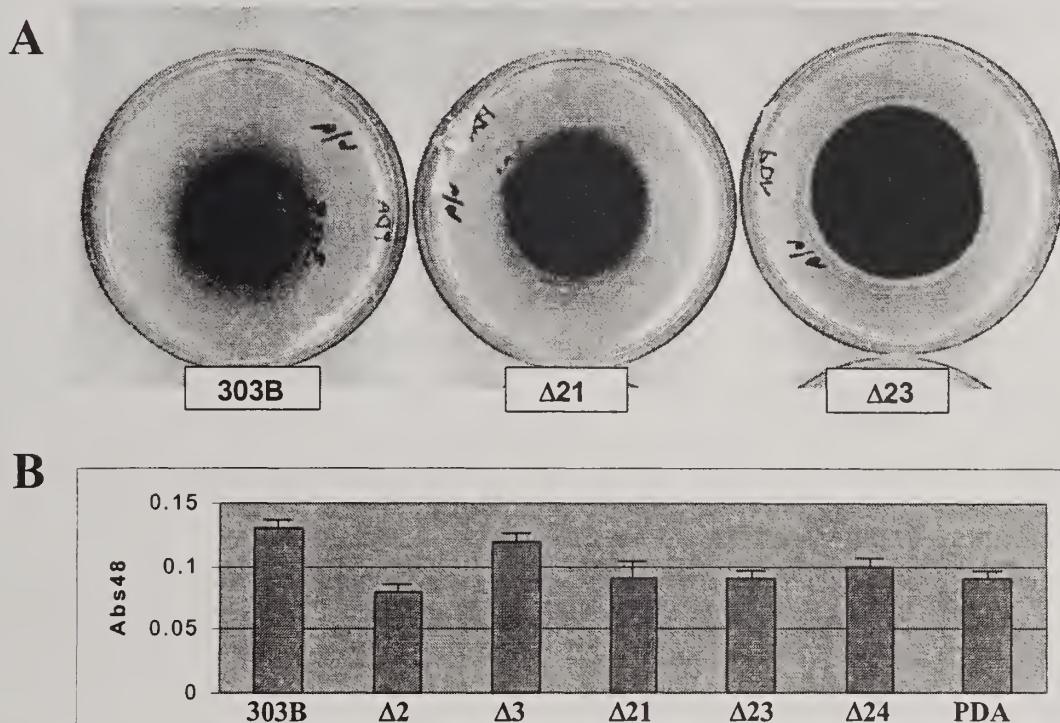
Figure 3.   Phenotype of *C. beticola* putatively lacking *ctb*1 gene expression. A. Parent isolate 303B was cultured in parallel with mutants *ctb*-Δ21 and Δ23 on PDA under continuous light. Note the red halo surrounding the 303B mycelium that is lacking or reduced in the transformants. B. Red pigment was eluted from the agar surrounding the mycelium and quantitated at 480 nm. Mean and standard deviation shown.

TAGGING OF GENES FOR DISEASE RESISTANCE IN SUGARBEET USING MOLECULAR GENETIC MARKERS
Project 622

John J. Weiland

Markers that tag regions of chromosomes that harbor genes contributing to disease resistance in sugarbeet can be of use in many aspects of research. Such landmarks on the genomic map can be used in marker-assisted selection in sugarbeet breeding programs. In addition the markers can provide information regarding the clustering or lack thereof regarding the distribution of resistance genes throughout the genome. Finally, chromosome markers can be integral tools in the identification of DNA clones that potentially harbor resistance gene sequences. Cloned resistance genes can be analysed for clues as to their mode of action and can be transferred between plant species using gene transfer technologies.

We have focused early efforts on the tagging of resistance to powdery mildew disease and to root knot nematode. Similar work has already been done in European laboratories the analysis of resistance to Cercospora leaf spot and Rhizomania diseases. Powdery mildew (*Erysiphe polygoni*) and root knot nematode (*Meloidogyne* spp) resistance in sugarbeet has recently been characterized by ARS colleagues in Salinas, CA. Both genes show promise for the genetic control of several races of the organisms causing these diseases. In collaboration with Drs. Robert Lewellen (ARS-Salinas) and J. Mitch McGrath (ARS-East Lansing), these resistance genes are being tagged using the random amplified polymorphism (RAPD) technique.

In efforts to produce more robust markers for the *Pm* gene conferring resistance to powdery mildew disease, advanced populations provided by Dr. Lewellen were screened in the greenhouse at Fargo in 2004. Sugarbeet populations CP04 and CP06 were the focus of gene tagging efforts following the preparation of DNA.

	resistant	susceptible	total	
Population CP05	96	13	109	Chi squared = 9.936
				P = 0.0016
Population CP03	86	19	105	Chi squared = 2.670
				P = 0.1023
Population CP06	81	29	110	Chi squared = 0.109
				P = 0.7412

Figure 4.   Powdery mildew resistance conferred by the *Pm* gene in sugarbeet: F2 segregation data of population CP03, 04, 05, and 06 at Fargo, 2004. F2 Populations from R.T. Lewellen (USDA-ARS, Salinas, CA) segregating for monogenic resistance to powdery mildew (Lewellen and Schrandt, 2001)

Susceptible Individuals **Resistant Individuals**
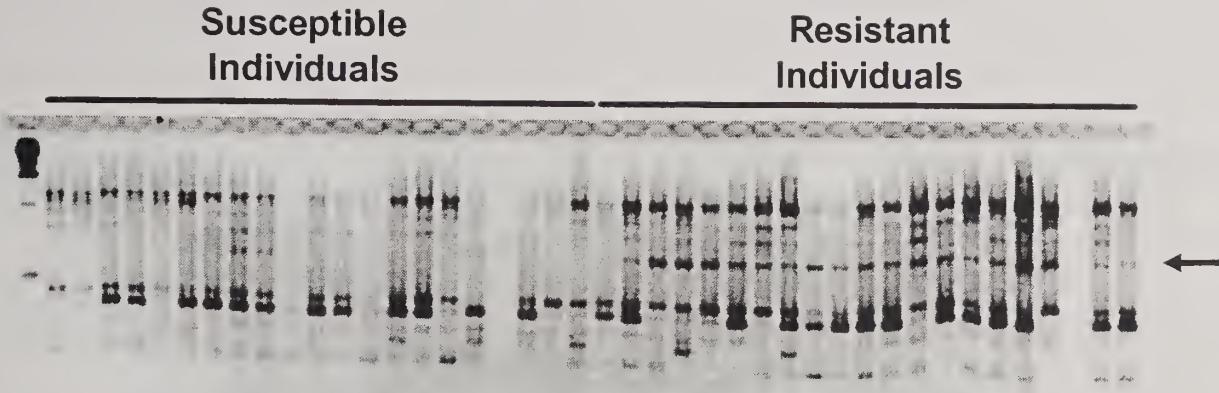
1.6% agarose gel of RAPD/Tbv products using decamer primers.

Figure 5. Powdery mildew resistance conferred by the *Pm* gene in sugarbeet: Tagging of segregants from CP06 population .

The marker shown in Figure 5 is presently being cloned and primers are being made for its exclusive detection by PCR. Should this marker prove to have high association with the Pm allele in all populations tested, the sequence of the marker will be made available to public and private breeders upon request.

Finally, studies on the segregation of resistance to *Aphanomyces* and sugarbeet cyst nematode (SBCN) will be done using populations generated by Dr. Bob Lewellen at USDA-ARS in Salinas, CA. Source materials from *B.vulgaris* spp *maritima* have yielded segregating populations with improved resistance to SBCN as assayed in California soils and under artificial inoculation. Discussions with Dr. Lewellen indicate that DNA should be able to be prepared by late 2004. Tagging of the gene(s) for resistance to this pest will commence shortly thereafter.

ROLE OF SUCROSE METABOLIZING ENZYMES IN SUGARBEET ROOT GROWTH, CARBOHYDRATE PARTITIONING AND POSTHARVEST SUCROSE LOSS
Project 650

Karen Klotz

Sucrose catabolism has been implicated as a major factor controlling whole plant carbon partitioning, root growth, sucrose accumulation, and postharvest sucrose loss (Wyse, 1974; Giaquinta, 1979; Sung et al., 1989; Zrenner et al., 1995; Berghall et al., 1997). In sugarbeet root, sucrose catabolism is catalyzed by three enzyme activities: sucrose synthase, acid invertase and alkaline invertase. Although all three activities are found in sugarbeet root, sucrose synthase is the predominant activity during root development and accounts for more than 90% of the total soluble sucrolytic activity during postharvest storage (Klotz and Finger, 2002; 2004). The enzyme is involved in sucrose utilization during development (Amor et al., 1995), has been implicated in sucrose partitioning to storage organs (Sung et al., 1989; Zrenner et al., 1995) and is believed to be involved in postharvest sucrose degradation (Echeverría and Gonzalez, 2003).

Two sucrose synthase proteins have been identified in sugarbeet roots (Klotz et al., 2003) and are encoded by two sucrose synthase genes (Hesse and Willmitzer, 1996; Haagenson et al., submitted). Previous research demonstrated that both genes were highly expressed in roots, but exhibited low to moderate expression in leaf and floral tissues. Research during the past twelve months examined the developmental expression of the two genes and the effect of environmental stresses on their expression. The objective of this research was to improve our understanding of sucrose synthase expression and the factors that control it, and to provide the molecular basis for future studies into sucrose synthase function in relation to root yield, sucrose accumulation and postharvest sucrose loss.

Developmental expression of sucrose synthase genes

Expression of the two sucrose synthase genes, sugarbeet sucrose synthase 1 (SBSS1) and sugarbeet sucrose synthase 2 (SBSS2), was determined during root development (Fig. 1). Transcript and protein levels of SBSS1 and SBSS2 were determined by Northern and Western analyses in roots of greenhouse-grown Beta 6225 at 3, 4, 5, 6, 8, 10, 12 and 16 weeks after planting. SBSS1 was expressed throughout development with transcript levels greatest during midseason growth (5 to 10 weeks after planting; Fig. 1A). In contrast, SBSS2 steady state mRNA levels were greatest during early development (3 to 6 weeks after planting), and were expressed at very low levels during late season development (12 to 16 weeks after planting). Developmental changes in SBSS1 and SBSS2 protein levels were similar to the changes observed for SBSS1 and SBSS2 transcript levels, although changes in protein levels were significantly delayed from transcriptional changes. The SBSS2 protein was predominant during early root development, equivalent to SBSS1 protein at 10 and 12 weeks, and present at slightly lower levels than SBSS1 at 16 weeks. The pattern of transcript and protein expression for the two genes suggests that expression may be largely regulated at the transcriptional level, but influenced by protein stability and other posttranscriptional mechanisms.

A

weeks:	3	4	5	6	8	10	12	16
SBSS1								
SBSS2								
25S								
18S								

B

weeks:	3	4	5	6	8	10	12	16
SBSS1								
SBSS2								

weeks	3	4	5	6	8	10	12	16
SBSS1	0	6	16	24	25	49	51	57
SBSS2	100	94	84	76	75	51	49	43
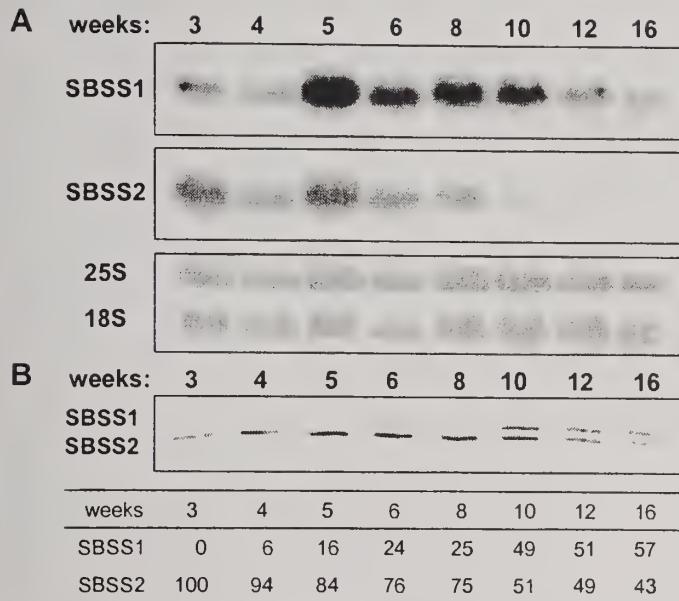
Figure 1: Developmental expression of sucrose synthase genes in root. (**A**) Northern hybridization (10 μg total RNA per lane) with ^{32}P-labeled SBSS1 and SBSS2 DNA probes, and methylene blue stained membrane showing RNA loading levels. (**B**) Western analysis (0.5 μg total protein per lane) using a sugarbeet sucrose synthase polyclonal antibody, and the relative abundance of SBSS1 and SBSS2 proteins as determined by densitometric scanning. Roots were harvested 3, 4, 5, 6, 8, 10, 12, and 16 weeks after planting.

Effect of environmental stresses on sucrose synthase gene expression

The effects of harvest, root injury, cold temperatures and anaerobic conditions on sucrose synthase expression were determined. In all studies, roots or plants of Beta 6225 were used. Plants were greenhouse grown in 15 L pots with supplemental light under a 16 h light/8 h dark regime and all experiments were initiated 16 weeks after date of sowing.

The effect of harvest and injury on sucrose synthase expression was determined by examining changes in SBSS1 and SBSS2 transcript levels, SBSS1 and SBSS2 protein levels, and sucrose synthase activity in harvested roots, with and without wounding, during storage for seven days at 20°C (Fig. 2). Roots were wounded by tumbling in a pilot lab beet washer for 30 minutes which caused surface abrasions, bruising and loss of the lower tail portion of the root. Unwounded (control) roots demonstrated the effect of harvest stress on expression. In these roots, transcript levels of both SBSS1 and SBSS2 declined (Fig. 2A). The decline in SBSS2 transcript was evident within 24 hours after harvest and persisted for seven days. The decline in SBSS1 transcript was not as great as that observed for SBSS2 and was only apparent after densitometric scanning of the Northern analyses and correction for loading differences using the intensity of the signal from the 18S ribosomal RNA (data not shown). Like SBSS2, SBSS1 transcript declined within 24 hours and remained depressed for seven days after harvest. Wounding caused a transient increase in SBSS1 and SBSS2 transcript. SBSS1 transcript was elevated during the first three days after injury, while SBSS2 transcript was elevated one day after injury. Despite changes in SBSS1 and SBSS2 transcript levels in response to harvest and wounding, no changes in SBSS1 and SBSS2 protein levels were apparent by Western analysis (Fig. 2B), although a transient increase in sucrose synthase activity of approximately 25 to 30% was observed in wounded roots two to three days after injury (Fig. 2C). The magnitude of the increase in activity contrasts with the two to three-fold increase in SBSS1 and SBSS2 transcript levels that occurred in response to wounding. The above experiment was repeated at 10°C. SBSS1 and SBSS2 transcript and protein levels exhibited similar changes at both storage temperatures (data not shown).

A

control   wounded

days: 0 1 2 3 4 7 1 2 3 4 7

SBSS1

SBSS2

25S

18S

B SBSS1
SBSS2

C

Figure 2: Effect of harvest and wounding on sucrose synthase gene expression. Roots were harvested 16 weeks after planting and incubated at 20°C and 90% relative humidity for up to 7 days. Wounded roots were severely bruised by tumbling in a pilot lab beet washer. (**A**) Northern hybridization (10 µg total RNA per lane) with ^{32}P-labeled SBSS1 and SBSS2 DNA probes, and methylene blue stained membrane showing RNA loading. (**B**) Western analysis (0.5 µg total protein per lane) using a sugarbeet sucrose synthase polyclonal antibody. (**C**) Sucrose synthase activity. Error bars are ± SE ($n = 4$).

Figure 3: Effect of cold on sucrose synthase gene expression. Roots were harvested 16 weeks after planting and incubated at 20°C for 3 days to allow for wound-healing and recovery from harvest. After 3 days, roots were incubated at 20 or 2°C for an additional 0.3 to 7 days. All incubations were at 90% relative humidity. (**A**) Northern hybridization (10 µg total RNA per lane) with ^{32}P-labeled SBSS1 and SBSS2 DNA probes, and methylene blue stained membrane showing RNA loading. (**B**) Western analysis (0.5 µg total protein per lane) using a sugarbeet sucrose synthase polyclonal antibody. (**C**) Sucrose synthase activity. Error bars are ± SE ($n = 4$).

A

20°C   2°C

days: 0 0.3 1 2 3 4 7 0.3 1 2 3 4 7

SBSS1

SBSS2

25S

18S

B SBSS1
SBSS2

C

The effect of cold temperature on sucrose synthase expression was determined by comparing SBSS1 and SBSS2 transcript and protein levels, and sucrose synthase activity in harvested roots stored at 20 and 2°C (Fig. 3). Roots were harvested and stored at 20°C and 90% relative humidity for three days to allow for wound-healing and recovery from harvest. At the end of three days, roots were incubated at 20°C or 2°C for up to seven days. SBSS2 transcript levels increased in cold treated roots beginning 3 days after the inception of the cold treatment (Fig. 3A). SBSS1 transcript levels, however, were unaffected by cold. Changes in SBSS1 transcript levels were apparent, but were related to time in storage, since similar changes were observed in roots stored at both temperatures. Despite changes in SBSS1 and SBSS2 transcript levels in response to temperature or time in storage, no changes in SBSS1 and SBSS2 protein levels were apparent by Western analysis (Fig. 3B), and sucrose synthase activity was unaltered by storage temperature (Fig. 3C).

The effect of anaerobic conditions on sucrose synthase expression was determined using intact plants to avoid the harvest-related transcriptional changes that were observed in previous experiments. Intact potted plants were submerged in water up to the apex of the taproot to create anaerobic conditions, and untreated control and anaerobic plants were maintained in the greenhouse for up to seven days. Anaerobic conditions caused SBSS1 transcript level to increase substantially after one day and led to a decline in SBSS2 transcript levels after two days (Fig. 4A). SBSS1 and SBSS2 protein levels (Fig. 4B), however, were relatively unchanged by anaerobic conditions. Sucrose synthase activity was unaltered by anaerobic conditions except for a transient increase in activity occurring after four days exposure to anaerobic conditions (Fig. 4C).

Figure 4: Effect of anaerobic conditions on sucrose synthase gene expression. Root tissue was harvested from intact potted plants after exposure to anaerobic conditions for up to 7 days. Anaerobic conditions were created by submerging plants in water up to the apex of the taproot. Plants were 16 weeks old at the initiation of the experiment. (A) Northern hybridization (10 µg total RNA per lane) with ^{32}P-labeled DNA probes for SBSS1, SBSS2, and 18S ribosomal RNA. (B) Western analysis (0.5 µg total protein per lane) using a sugarbeet sucrose synthase polyclonal antibody. (C) Sucrose synthase activity. Error bars are ± SE ($n = 4$).
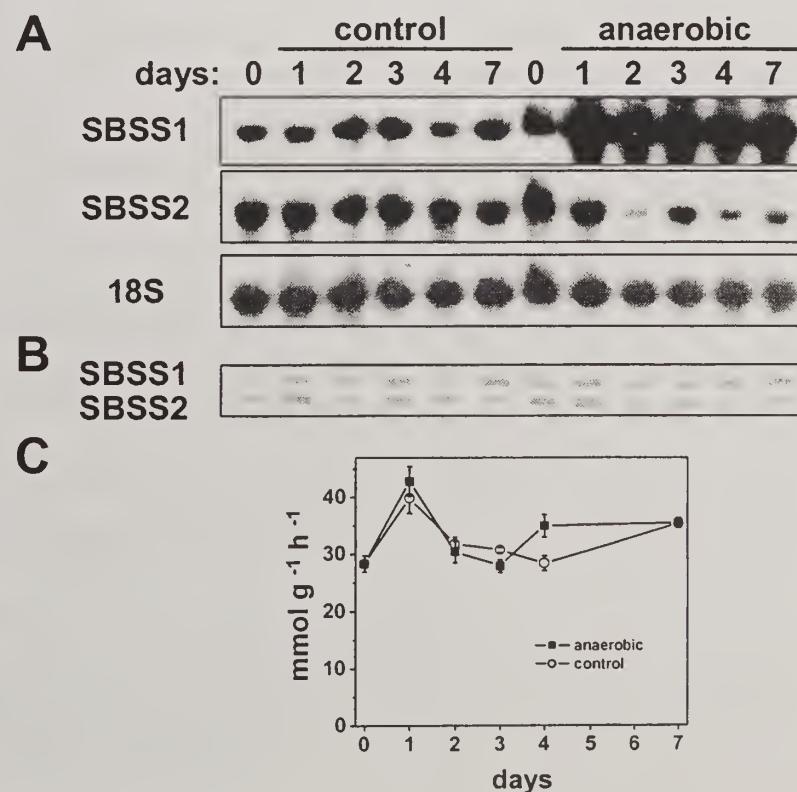
Conclusions

➤ The two sucrose synthase genes are developmentally expressed in roots, with SBSS2 expression greater during early development and SBSS1 expression greater during late season development. Comparison of transcript and protein expression of sucrose synthase genes during development suggests that regulation of expression is not strictly transcriptional. Sucrose synthase activity is likely to be influenced by protein stability, and possibly other posttranscriptional mechanisms.

➤ Sucrose synthase activity was largely unresponsive to harvest, injury, cold temperatures and anaerobic conditions. These environmental stresses altered sucrose synthase transcript levels but had little or no effect on sucrose synthase protein levels or activity, suggesting that regulation of sucrose synthase expression in response to environmental stresses is primarily posttranscriptional.

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CHARACTERIZATION OF RAFFINOSE BIOSYNTHESIS DURING SUGARBEET PRODUCTION AND STORAGE
Project 651

Karen L. Klotz

Raffinose is a carbohydrate impurity that interferes with sugarbeet processing by decreasing the yield of extractable sucrose and altering sucrose crystal morphology resulting in slower filtration rates and slower processing. Although low, nonfreezing temperatures are closely associated with increased raffinose concentrations during sugarbeet growth and storage (Wyse and Dexter, 1971), the physiological and biochemical mechanisms associated with raffinose accumulation are poorly understood. In other plant species, the raffinose biosynthetic pathway has been characterized (Peterbauer and Richter, 2001). Raffinose synthesis is catalyzed by raffinose synthase, an enzyme that transfers a galactosyl unit from galactinol to sucrose. The formation of galactinol is the first committed step in the synthesis of raffinose and this reaction is catalyzed by galactinol synthase. The objective of this research was to evaluate sugarbeet raffinose accumulation, raffinose biosynthetic gene expression, and raffinose synthase enzyme activity during production and storage in an attempt to identify the physiological, biochemical, and environmental factors associated with sugarbeet raffinose accumulation.

In 2004, a galactinol synthase cDNA was obtained from a sugarbeet EST collection (KWS, Germany), and a sugarbeet-specific raffinose synthase cDNA clone was isolated by PCR. Methods to evaluate raffinose synthase enzyme activity were adapted from an established assay in soybean (Hitz et al., 2002).

Harvest Date, Storage Temperature and Storage Duration
The first year of a postharvest storage study examining harvest date, storage temperature and storage duration on raffinose accumulation, raffinose biosynthetic gene expression, and raffinose synthase enzyme activity was completed in 2004. Field-grown beets from a Fargo, ND, location were sampled at three harvest dates (8 September, 23 September, and 29 October, 2003), and beets were stored for 2, 10, and 18 weeks at 2 or 6°C.

Raffinose accumulation and raffinose synthase activity. Storage trends in raffinose accumulation and raffinose synthase activity were similar among all harvest dates and data was pooled across harvest date (Figure 1). When averaged across storage duration, raffinose concentrations were three-fold higher in roots stored at 2°C than in roots stored at 6°C, and raffinose synthase activity was two-fold higher in roots stored at 2°C than in roots stored at 6°C. Beets stored at 2°C had a significant increase in enzyme activity after 2 weeks in storage with a concomitant rise in raffinose concentrations. Enzyme activity from beets stored at 2°C remained elevated throughout 18 wk of storage, while raffinose synthase activity from beets stored at 6°C decreased slightly throughout storage. After 10 weeks of storage, there was a four-fold increase in raffinose concentration in beets stored at 2°C, and concentrations remained elevated through 18 wk of storage. In contrast, beets stored at 6°C had a small increase in raffinose after 10 wk in storage, and raffinose concentrations declined by 18 wk to initial storage (0 wk) levels.
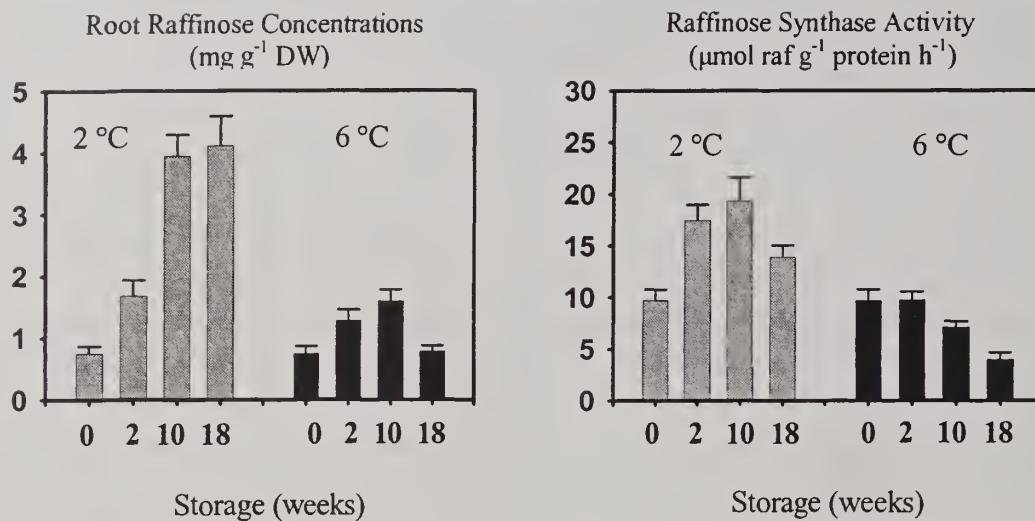
Figure 1: Impact of storage temperature and duration on root raffinose concentration and raffinose synthase enzyme activity. Data are the mean ± SE of 12 replicates.

Raffinose biosynthetic gene expression. Beets stored at 2°C had higher galactinol and raffinose synthase steady-state mRNA levels when compared to beets stored at 6°C (Figure 2). Raffinose biosynthetic gene expression was highest in roots stored for 2 wk at 2°C and mRNA transcript abundance decreased markedly at 10 and 18 weeks of storage for both storage temperatures. Raffinose synthase and galactinol synthase transcript levels were similar among the three harvest dates at all storage conditions except for galactinol synthase at time 0 where transcript abundance was highest in beets harvested 29 October. The increased galactinol synthase expression in late October may be associated with decreased temperature in October when compared with beets sampled in early and mid-September.
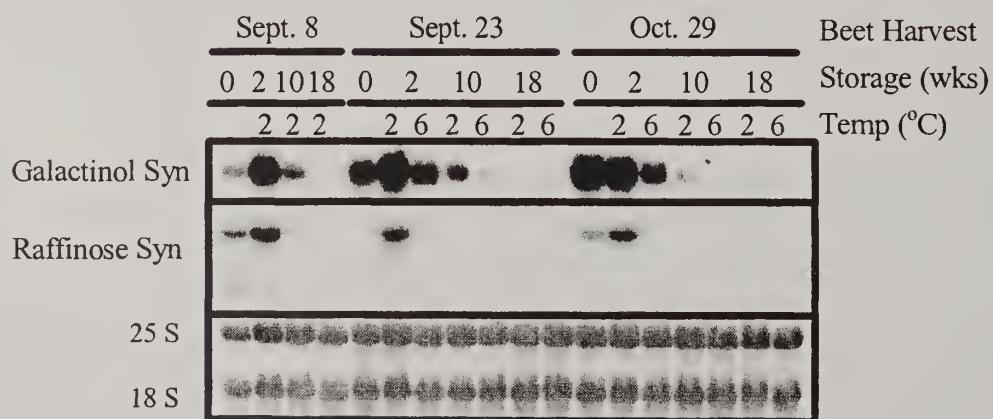
Figure 2: RNA blot analysis of raffinose biosynthetic gene expression in storage. Total RNA (10 μg per lane) was hybridized with sugarbeet galactinol synthase or raffinose synthase cDNA clones. The lower panel is the methylene blue stained nylon membrane showing ribosomal RNA levels.

Influence of desiccation stress on raffinose accumulation.

Harvested roots from two commercial sugarbeet varieties (Beta 6225 and VDH 66156) were stored in 50 and 90% relative humidity (RH) cabinets at 10°C to examine the impact of desiccation stress on raffinose accumulation and raffinose synthase enzyme activity. Both varieties had a similar response in raffinose accumulation and raffinose synthase enzyme activity under desiccation, and the reported data is the average of the two varieties (Figure 3). After four weeks of storage, there was a ten-fold increase in raffinose synthase activity from beets stored at 50% RH. In contrast, there was only a two-fold increase in raffinose synthase enzyme activity from beets stored at 90% RH after four weeks of storage. Surprisingly, raffinose accumulation did not correspond to raffinose synthase activity. Raffinose did not accumulate with desiccation stress (50% RH storage conditions), but instead, a six-fold increase in raffinose accumulation was measured after four weeks of storage at 90% RH. The physiological explanation for this fluctuation between raffinose accumulation and enzyme activity under desiccating environments is not known, but the impact of desiccation on raffinose-degrading galactosidase enzyme activity is being evaluated.
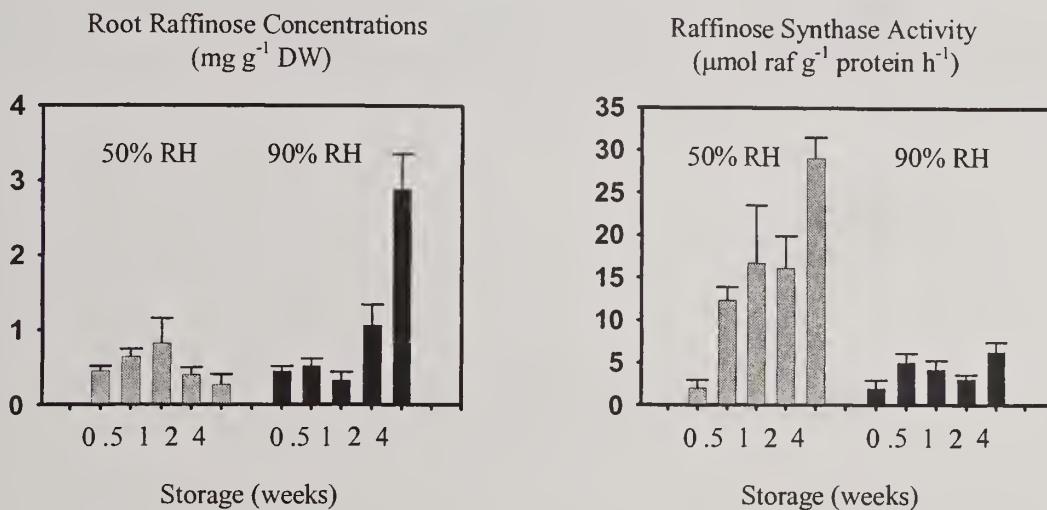
Figure 3: Impact of desiccation on root raffinose concentration and raffinose synthase enzyme activity. Data are the mean ± SE of 8 replicates.

Conclusions:

- Raffinose accumulated during sugarbeet storage, and decreased temperature (2°C) was associated with increased raffinose concentration, raffinose synthase enzyme activity, and transcript abundance.

- Raffinose accumulation and raffinose synthase enzyme activities were similar among the three harvest dates at all storage conditions in this first year of a two-year field study.

- Enzyme activities increased at 2 wk of storage from beets stored at 2°C. Although transcript levels decreased in late storage, enzyme activity was maintained at 10 and 18 wk of storage.

- Differences between transcript abundance and protein activity suggest that protein stability may have an important role regulating raffinose accumulation during sugarbeet storage.

- Desiccation stress increased enzyme activity, but not raffinose concentrations.

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SUGARBEET RESEARCH

2004 REPORT

Section D

Sugarbeet and Bean Research Unit
Agricultural Research Service – USDA
East Lansing, Michigan

Dr. J. M. McGrath, Sugarbeet Geneticist

Table of Contents

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Manuscripts published

Trebbi, D., McGrath, J.M. (2004) Fluorometric sucrose evaluation for sugar beet. J. Agricultural and Food Chemistry 52: 6862-6867.

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Introduction

The Sugarbeet and Bean Research Unit at East Lansing, MI has projects involved with sugarbeet, dry bean, apple, and cucumber. Two positions are open, a sugarbeet pathologist and a dry bean geneticist, and these will be recruited in the coming months. The sugarbeet program has three primary areas of investigation. First is breeding enhanced germplasm for adaptation to the Eastern US growing areas, with a priority on high sucrose, smooth root, and seedling disease resistance. Second is determining genetics of agronomic traits including sucrose accumulation, inheritance of seedling disease resistance, developing recombinant inbred lines, and constructing and characterizing molecular tools for the community (genetic maps, expressed sequence tags, bacterial artificial chromosome libraries). Third is the investigation of seedling vigor, including field emergence and stand establishment, stand persistence, development of *in vitro* germination and vigor tests, and molecular characterization of early plant development.

Sugar beet activities of the USDA-ARS East Lansing conducted in cooperation with Saginaw Valley Bean and Beet Farm during 2004

J. Mitchell McGrath, Tim M. Duckert, Teresa Koppin, Scott Shaw, and Daniele Trebbi

Five evaluation plots were planted at the Saginaw Valley Bean and Beet Research Farm in 2004; three agronomic and two disease nursery trials. All seed planted was untreated to maximize stand and seedling vigor traits inherent in the breeding germplasm. Two agronomic trials (04BB01 and 04BB02) were planted into Range 3, following normal fall tillage and seedbed preparations, on April 14, 2004. The remaining tests were planted on June 14. Blocking and thinning was completed by June 18. Harvest was October 12, and sucrose determinations were done on brei samples taken two days later, frozen, and sent to Seedex, Inc. for analyses. The contributions of Seedex and Michigan Sugar are gratefully acknowledged.

Test 04BB01: This test was conducted to evaluate first generation populations derived from field increases of (1) high sucrose, smooth-root materials (SR96 & SR97) inter-pollinated with EL0204, (2) experimental hybrids of SR94 and SR97 from CMS crossing blocks in Oregon, (3) inter-pollinated populations derived from viable long-term seed stored at East Lansing since 1985, (4) smooth-root germplasm with two cycles of selection from the Rhizoctonia nursery in East Lansing, (5) Cercospora resistant reselections of EL50 performed in the 2003 Cercospora nursery at the Bean and Beet farm, (6) seed selected from very long term seed storage at East Lansing (pre-1970), and (7) an over-wintered field crossing block at East Lansing; along with check entries (Table 1, purpose numbers refer to these headings). This experiment was single 24' rows with four replications in a randomized complete block design.

In general, performance was excellent. All entries showed good to excellent emergence, except Entry 3 where seed quality was very low and this entry failed to give a harvestable stand (Table 2). New for this year is an assessment of laboratory germination under stress (150mM NaCl) and non-stress (H$_2$O$_2$) liquid germination. These treatments have been used in the laboratory to deduce some molecular events associated with expression of seedling vigor. Ideally, a measure of Field Emergence Potential based on these or similar results will result with a full multi-year dataset.

The commercial check Betaseed B5736 outperformed all other entries, however this was not statistically different from Syngenta (Hilleshog) E17, SR94, and 15 experimental entries (Table

1). Experimental entries were predominantly first generation, open pollinated, field seed increases from selected mother roots of recently released East Lansing ARS germplasm conducted to effect inter-pollination to recombine high sucrose, smooth root, and disease resistance (particularly Holly-gene rhizomania resistance) into improved populations. At least one further cycle of selection and recombination will be done in 2005 in order to introgress traits into planned releases for 2007. Sites of field increases are abbreviated in the prefix of the entry (OS-Old Soils Farm; MF-Muck Farm; OB-Old Botany; BI-Botany Irrigation; RA-Range A; OW-Over Winter nursery). Other abbreviations used include smr (selected mother roots), IP (inter-pollinated), and HTLTS for High Temperature Long Term Storage of legacy germplasm from the late 1960's as a potential source of unique seed storability traits.

One observation of note is evident in Table 3. Water content (as a proportion of total root weight) was included in the analyses for the first time this year, and did not vary greatly among any germplasm, however differences are highly precise and robust. Noteworthy is that the commercial germplasms B5736 (entry 42) and E17 (entry 41) had at least 1% reduced water content relative to all experimental lines and checks. The significance of this observation is not yet entirely clear, but higher dry matter (DM) content appears to be a character amenable to selection, and does a higher ratio of sucrose as a proportion of DM (Suc/DM). Suc/DM also appears to vary among these populations, suggesting selecting for this criterion could result in further genetic gains. Other information in Table 3 includes sucrose content predictions using novel techniques we are developing such as Enzymatic-Fluorometric Analysis (EFA) for laboratory and Near-Infra Red (NIR) analyses for real-time sucrose selection at time of harvest. Both methods show promise.

Test 04BB02: This test was conducted to evaluate entries for possible inclusion into the germplasm release stream. Entries (listed in Table 4) were similar to those in Test 04BB01 except their predicted performance was generally expected to be inferior based on the performance of the parents. The test was conducted in three non-random replicates of 24' rows each, and this arrangement was particularly useful in discriminating morphological and agronomic characters such as Rhizoctonia reaction, where Entries 11 and 40 appeared particularly susceptible. Sucrose data was not taken for this test, but yield was measured. Entries exceeding 19 T/A will be tested in the 2005 agronomic nursery (where seed is available) for expanded agronomic analyses, and germplasm released if performance meets expectations. Additional information in Table 4 reflects the average plot weight in pounds, the average number of beets harvested per entry, and both 10- and 20-day stand counts. Low stand counts lead to low plot weights, reinforcing the need for good stands early in the season.

Test 04BB05: This test was done to evaluate performance of inbred lines of beets, in this case derived from a cross between sugar beet and red beet and derived through single seed descent to the F_5 with selection for only green plant segregates in the F_4, as a tool in genetic analyses. Inbreds often show reduced vigor relative to hybrids, and this was the case here, but genetic analyses are hampered by the out-crossing, hybrid nature of the crop. The use of inbred lines will help to deduce the genetic component of traits, and help in assigning observed variability into genetic and environmental components, something that has not been previously possible. The test was designed as a single replication of each entry in a 24' plot. Stand was good, despite being planted late. At harvest, five beets per plot were harvested by hand after machine lifting, weighed individually, and scanned with NIR to estimate yield components of sucrose content and water content, from which dry matter and sucrose as a proportion of dry matter were

calculated (Table 5). Significant differences were observed between entries for all characters, suggesting that their genetic components can be identified through this approach. Some lines showed similar beets weights as the commercial check (e.g. entry 94), suggesting that inbreeding *per se* is not always detrimental.

Test 04BB04: This test was done to evaluate germplasm for Cercospora resistance. Both Cercospora and Powdery mildew pressure were high in this nursery. The predominant entries in this test were from F_2 populations of crosses between C869 as a self-fertile seed parent and EL50 as a Cercospora resistance pollen parent. The test was primarily designed to choose the better F_2 families for developing recombinant inbred lines that will continue in the greenhouse. No agronomic performance measures were done, but the task of selecting germplasm to advance to the next generation was successful.

Test 04BB03: This test was done as a continuing project to select for survival and growth of beets in the agronomically depauperate soils north of the farm pond. A wide mix of germplasm was grown, including advanced generation materials from a cross between sugar beet and a wild beet (PI540625) with reported high levels of resistance to Aphanomyces. Stand counts were the major data collected (Table 6), with some notes recorded on disease severity. Disease severity on roots was not pronounced at harvest and only varied by a small degree between entries. One entry (designated Hero) is the most advanced of the materials with the PI540625 background, and will be released as germplasm in 2005. It should be noted that the disease spectrum in this nursery is broad, and includes Aphanomyces, Rhizoctonia, and Pythium that all can serious affect stand persistence.

Self-fertility in beets: Obligate or facultative?

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Beets are normally out-crossing, enforced by a complex system of self-incompatibility, asynchronous flowering, and different times of maturation of pollen and stigma in the same flower. The Self-fertile trait (Sf), a monogenic dominant character, can overcome self-sterility. This character is very useful in developing defined segregating populations for genetic analyses. It is not clear whether Sf is an allele at a self-incompatibility locus, or a supressor of incompatibility. Neither Sf nor the incompatibility loci have been genetically mapped.

For practical reasons during routine selfing of individuals in populations, it would be useful to know if selfing would happen in a mixed pollen environment. That is, is it necessary to individually isolate flowers and racemes (e.g. bagging) or is it possible to obtain true breeding individuals without isolating them from other pollen sources? Bagging plants represents a significant labor cost that could be avoided if genetic unnecessary.

We have been doing single seed decent on a population derived from a cross between self-fertile sugar and red table beets, and had the opportunity to test for the strength of the Sf trait by examining seed harvested from green male-sterile (nuclear) and male-fertile plants when provided with a red beet pollinator. The monogenic Red trait (R) is dominant to green (rr), thus any seed with intensely red hypocotyls harvested from a green plant were due to out-crossing, and thus the rate of cross pollination could be estimated.

Plants (in 4 l pots) were grown in Greenhouse 31B, which is ca. 8 m (east-west) x 3 m (north-south) and equipped with two 1 m exhaust fans and two 300 cm circulating fans providing

continuous air movement. Exhaust fans operated continuously for 12 hours per day during daylight hours. Four red beet plants were placed 1 m from the west wall and equi-distant from the wall in the north-south orientation. Green plants were placed at 6.0 m laterally from the red beets (e.g. a bench on the opposite side of the room), or on the same bench as the red beets at both 2.0 and 0.5 m distant. Proximity to the exhaust fans was recorded as either upwind (away from the fans) or downwind (close to the fan, ca. 0.5 m distant). Each male sterile green plant was paired with a fertile plant at the same location but not bagged. In addition, three pairs of bagged crosses using one green fertile plant and one red fertile plant were placed in the center of the room.

Results indicated that self-fertility is not obligate (Table 7). As expected, seed harvested from bagged red beets paired with green beets showed all red hypocotyl. Out-crossing as determined from the proportion of red hypocotyl seedlings from seed harvested from the green plant of the pair-cross averaged 8.8%. This represents the level of contamination that could be expected due to proximity and contact of flowers of different self-fertile plants (inbreds), and likely was a consequence of normal plant care activities such as watering, fertilizing, and pest control. Airflow had an unpredictable influence on pollen contamination, however generalizations of separation by distance and direction of the prevailing wind seemed to reduce contamination. Some of the uncertainty was caused by inclusion of a green pollen source near each sterile plant and this undoubtedly reduced the number of red hypocotyl seedlings on male sterile plants. However, we were also interested in the proportion of red contamination in green self-fertile pollinators, and this value averaged 7.3% but with high variability. In conclusion, the self-fertility trait is not absolute, and allows an average of ca. 10% contamination if plants are not properly isolated.

Table 7: Proportion of red hypocotyl seedlings from self-compatible green parents inter-pollinated with red beet.

Plant #	Plant Color	Fertility	Pollination	Distance (m)	Airflow	Hypocotyl Red	Green	Total	% Red
03B117-1	red	fertile	bagged	0.0	none	35	0	35	100.0
03B182-2	red	fertile	bagged	0.0	none	39	0	39	100.0
03B224-1	red	fertile	bagged	0.0	none	30	0	30	100.0
03B179-1	green	fertile	bagged	0.0	none	18	186	204	8.8
03B158-1	green	fertile	bagged	0.0	none	17	181	198	8.6
03B141-3	green	fertile	bagged	0.0	none	17	171	188	9.0
03B192-2	green	fertile	open	6.0	downwind	1	34	35	2.9
03B166-3	green	fertile	open	2.0	downwind	4	17	21	19.0
03B118-4	green	fertile	open	0.5	downwind	9	79	88	10.2
03B166-1	green	fertile	open	6.0	upwind	6	61	67	9.0
03B149-1	green	fertile	open	2.0	upwind	0	58	58	0.0
03B153-1	green	fertile	open	0.5	upwind	2	79	81	2.5
03B124-3	green	sterile	open	2.0	downwind	13	38	51	25.5
03B121-1	green	sterile	open	0.5	downwind	1	0	1	100.0
03B191-3	green	sterile	open	6.0	upwind	5	25	30	16.7
03B135-1	green	sterile	open	2.0	upwind	12	111	123	9.8
03B191-1	green	sterile	open	0.5	upwind	9	20	29	31.0

Sugar beet seedling disease resistance caused by Rhizoctonia.

Subashini Nagendran and J. Mitchell McGrath (*BSDF Project 742*)

Early season growth (e.g. the first 10 weeks) is a critical phase of the beet's life, not only to have good field stands but also to acquire metabolic capacity for agronomic productivity. Early season development includes acquisition of disease tolerance (from acute symptoms with devastating effects to chronic symptoms that only reduce yield potential), and development of the taproot. This change from seedling to adult vegetative growth coincides, in the field, with warming temperatures (and greater seedling disease), increased growth rate, and increased light interception. Yield of sucrose is directly proportional to the interception of solar irradiation, and maximal interception of sunlight does not occur until the crop canopy is fully developed usually past the summer solstice. Most (if not all) constructive agronomic processes are in place by the 10th week after emergence. Disease losses are a constant concern through the growing season and during post-harvest storage, but are caused by a relatively small number of pathogens for which genetic resistance is generally available, however seedling disease and competition from weeds have the greatest impact on obtaining a profitable crop. The focus of this project has been to evaluate the host-pathogen interaction between sugar beet seedlings and *Rhizoctonia solani* with the aim of discovering mechanisms of resistance, and apply this in breeding for enhanced stand persistence.

Rhizoctonia diseases are increasingly important in the Great Lakes growing region, and elsewhere. Genetic resistance is available for the chronic phase of the disease (crown and root rot), and a number of germplasm lines have been released over the past 20 years, which are now becoming available as resistant hybrids available through seed companies. Seedling resistance to Rhizoctonia blight has only recently been reported. Both crown and root rot and seedling diseases are caused by *Rhizoctonia solani*, a biologically complex species with many sub-types (Anastomosis Groups, AG), of which AG2-2 is the most serious to sugarbeet, and AG4 has been implicated as a pathogen only during seedling growth.

When seedling hypocotyls are infected at or below ground level, diseased seedlings collapse and die (e.g. damp-off). This leads to lack of full stand persistence and is implicated in the range of problems associated with emergence and stand establishment encountered by growers worldwide. Early-season Rhizoctonia disease research has been stimulated with an observation that Quadris and Amistar, fungicides with anti-Rhizoctonia activity, applied early in the season can significantly increase harvest yields. While the precise mechanism is unclear, this observation supports objectives for improving seedling emergence and stand establishment, and suggests that one of the major biotic impediments to improving stand persistence is, indeed, seedling disease caused by Rhizoctonia.

Little has been published on the topic of Rhizoctonia seedling resistance in sugarbeet, although the chronic crown and root rot phase of the disease in sugarbeet has received considerable attention. The primary invasion sites for crown and root rot are lower surfaces of petioles in contact with the soil, natural cracks in the crown, lenticels on the taproot, lateral roots, and opportunistic secondary infections after damage by nematodes or other penetrations. Rhizoctonia seedling diseases of sugar beet differ in pathogenicity and virulence from those causing root rot on older beets. The mode of penetration and the progress of subsequent tissue colonization play important roles in Rhizoctonia causing diseases. There is no reported resistance to seedling disease caused by Rhizoctonia.

Materials and Methods

Fungal inocula: *R. solani* fungal isolates were grown on Corn Meal Agar (CMA; Criterion, Hardy diagnostics, C5491) in Petri dish at room temperature. De-hulled seeds of millet, sterilized on three consecutive days at 120°C for 20 minutes each day, were placed as single layer on the actively growing 3 day old CMA fungal culture and were incubated at room temperature in the light for an additional four days. The millet was completely colonized with the fungus, and this was used as the inoculum.

Bioassays of virulence –Growth chamber disease screening protocol: USH20 was used to develop the screening protocol. Pots (9 cm diameter by 8 cm deep) on cafeteria trays were filled to 2 cm below the top with "Baccto" high porosity soil and were arranged in a randomized complete block design. Four germinated seeds were planted per pot and grown in a growth chamber (20°C, 20 hour light and 4 hour dark photoperiod), watered daily, fertilized weekly, and thinned to three plants for the test. Seedlings at the 4 to 6 leaf-stage were inoculated with a single *R. solani* isolate, with five pots (15 plants total) inoculated per isolate.

Isolates of *R. solani* isolated from sugar beets were used, (kindly provided by Dr. Lee Panella and Dr. Linda Hansen, USDA-ARS, Ft. Collins, CO), one each of a virulent (strain R1) and avirulent strain of AG2-2 (strain W2). AG4 isolates were also tested. The amount of inocula to be used per plant to infect them was optimized by taking into consideration that seedlings should not be killed rapidly and damping off symptoms should progress gradually. Sugar beet seedlings with 4 to 6 leaf stage were inoculated with fungus colonized millet seeds. Each seedling was inoculated by surrounding each plant with 10 fungus-infected millet seeds, 2 cm from each seedling. Control plants were inoculated with sterile millet. Post inoculation observations were made at one day intervals (Post inoculation days, PID) and the symptoms were recorded (0=healthy; 1=slight penetration scar visible to naked eye; 2=deep penetration scar very visible, margin of the wound brown to black color; 3= plant showing damping off symptoms, petioles loosing turgor and rigidity, hypocotyls (stem) shows water soaked lesions; 4=plant damping off, leaf blades wilting; and 5=plant dead. Fifteen seedlings per treatment (fungal strain) were scored and the average score is reported as disease index (DI).

Rhizoctonia seedling damping-off disease progress pattern in the greenhouse: Wooden boxes (400 x 580 cm) were filled to 2 cm below the top with a high porosity soilless mix (Baccto, perlite and peat) and were arranged in a randomized complete block design. Thirty germinated seeds were planted per wooden box and grown in the green house (25°C, 16 hr light and 8 hr dark photoperiod), watered daily, fertilized weekly, and transplanted and maintained at thirty plants per box. Seedlings with 4 to 6 leaf stage were inoculated with a single isolate of *R. solani* AG 2-2, R1 (virulent) strain or W2-2 (avirulent) strain. Each seedling was inoculated by adding 0.1 g of inocula (about 20 fungus –infested millet seeds.) on 2 opposite sides of each plant 4 cm away from each seedling. Control plants were inoculated with uninfected, sterile millet. Rhizoctonia seedling damping off disease progress was scored at daily intervals. Forty seedlings per treatment (fungal strain) were scored and the average score is reported as disease index (DI). Ten seedlings per treatment- box were used to make microscopic observations (data not shown) and isolate pathogen as the disease progresses. Each treatment was repeated. The experiment was repeated under varying external seasonal conditions in the green house.

Rhizoctonia seedling damping-off disease progress pattern in the field: Field experiments in East Lansing MI were established in 2003, with plots arranged to separate treatments with

different strains of *R. solani*. Four sugar beet germplasm lines were tested (USH20; 00B041 - a Hogaboam-era collection of crown and root rot resistant selections; 01B024 – a smooth root breeding line twice selected for crown and root rot resistance; and EL51 - bred for resistance to Rhizoctonia root and crown rot). Treatments were *R. solani* AG2-2 R1 (virulent strain), AG2-2 W2-2 (avirulent), AG2-2 Michigan 7201 (virulent, positive control), and a negative control inoculated with sterile millet seeds. At the 6- to 8- leaf stage (about 4 weeks after sowing or 2 weeks after germination) plants were manually thinned to 10 cm spacing. Following at least a three day recovery, each seedling was then inoculated by adding about 3.3 g of inocula on one side of each plant 4 cm away from each seedling. The incidence and the development of *R. solani*-induced sugar beet seedling damping-off were assessed by counting the number of emerged seedlings (stand count) in each plot 10, 12, 14, 20, and 25 days after inoculation. The experiment was repeated in 2004, using only USH20 and EL51.

Results and Discussion

The disease progress curves showed three phases of disease reaction on USH20. The initial infection (from zero to 9 days post-inoculation) was characterized by rapid appearance of symptoms, the second phase (from 8 to 13 days of post-inoculation) was characterized by little disease progression, and the final phase (14 to 18 days post-inoculation) finalized the outcome of the interaction, either death (compatible interaction) or recovery (incompatible interaction) (Figure 1). Virulent AG2-2 (strain R1) caused seedling blight and death of the plant. Seedlings infected with three other isolates (AG2-2 strain W2, and AG4 strains R6 and R7) showed fewer blight symptoms and less mortality than with AG2-2. At post inoculation day (PID) 6, disease severity actually decreased for a period of 1 to 2 days before rising and reaching a plateau by PID 9, but only for AG4 and the avirulent isolate of AG2-2 (i.e. isolate W2). For virulent isolate AG2-2 R1, this same time frame was characterized by a rapid increase in disease severity, followed by a plateau. By PID 14, all plants infected with the virulent isolate were dead, while avirulent AG2-2 (i.e. isolate W2) and each of the AG4 isolates had begun to recover and were showing limited symptoms.

Figure 1: Disease interaction between USH20 and virulent and avirulent *Rhizoctonia solani* isolates determined in the greenhouse (growth chamber results were similar).
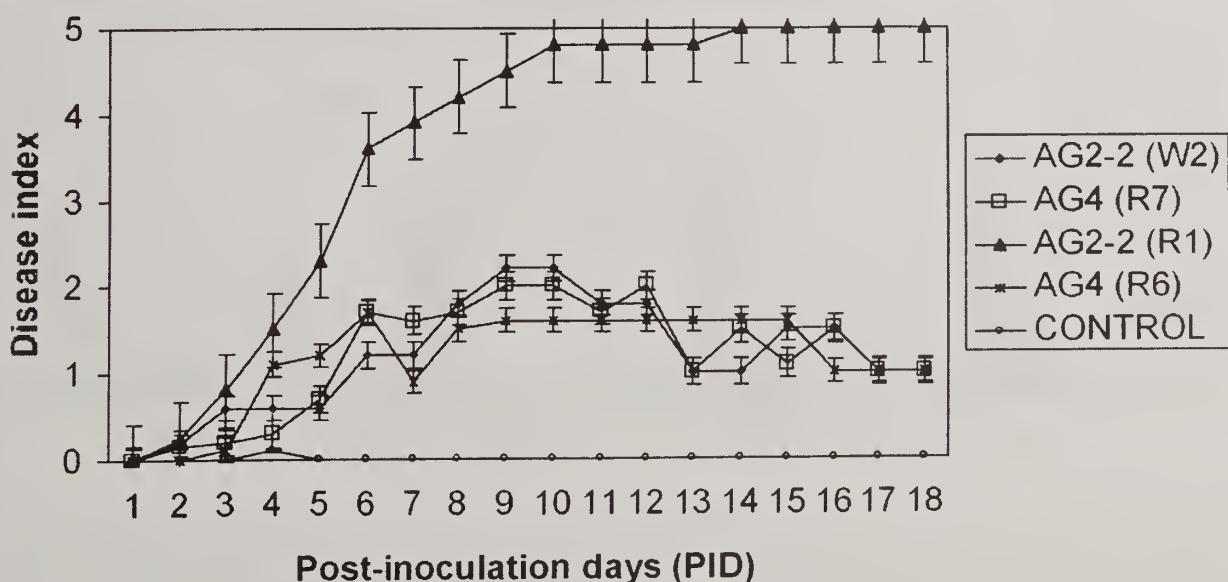
AG2-2 was more damaging to sugar beet seedlings in general. Neither the *in vitro* saprophytic growth rate nor in vitro sclerotium producing ability showed any correlation with the fungal pathogenesis on sugar beet seedlings. Observations indicated that *R. solani* avirulent strains initiated infection but failed to establish and cause disease. Separate results indicated prior infection by avirulent strains did not induce host resistance.

In the greenhouse, different accessions of sugar beet were tested for resistance to Rhizoctonia seedling blight. Plant material consisted of different releases of sugar beet (*Beta vulgaris*) with different levels of resistant to various diseases and other beet quality traits and wild accessions (*B. v.* ssp. *maritima*). Sugarbeet EL51 showed partial resistance to seedling damping off caused by AG2-2 (R1). The sugar beet accessions that were reported to be resistant to Rhizoctonia crown and root rot were susceptible to Rhizoctonia seedling blight suggesting that there is little or no correlation between these two disease reactions.

Rhizoctonia seedling blight in the field was analyzed in 2003, with plots arranged to separate treatments with different strains of *R. solani* at the Botany Farm in East Lansing, Michigan. Seven replicates, each 20' long containing from 30-50 plants of four germplasm lines (USH20, 00B041, 01B024, and EL51). At the 6 - 8 leaf stage (about 2 weeks after emergence), each seedling was inoculated with ca. 3.3 g of inocula on one side of each plant, 4 cm away from each seedling. The incidence and development of *R. solani*- induced sugar beet seedling blight was assessed by counting the number of emerged seedlings (stand count) in each plot at 21 and 35 days after inoculation. Symptoms of *R. solani*-induced blight were observed on all sampled seedlings (i.e. 10 % of wilted seedlings were harvested and examined). The number of dead plants was low in sterile millet treatments and in treatment plots inoculated with avirulent compared to that of virulent strains (Figure 2). Virulent isolate R1 caused the greatest mortality, however little mortality was seen with EL51. Final stand counts were more dramatic, with only EL51 showing a high final stand count.

Figure 2: Rhizoctonia seedling damping off in the 2003 field at 21 days post-inoculation, as compared to control. (-) ve control was sterile millet inoculated seedlings, (+) ve control was inoculated with virulent isolate 7201.
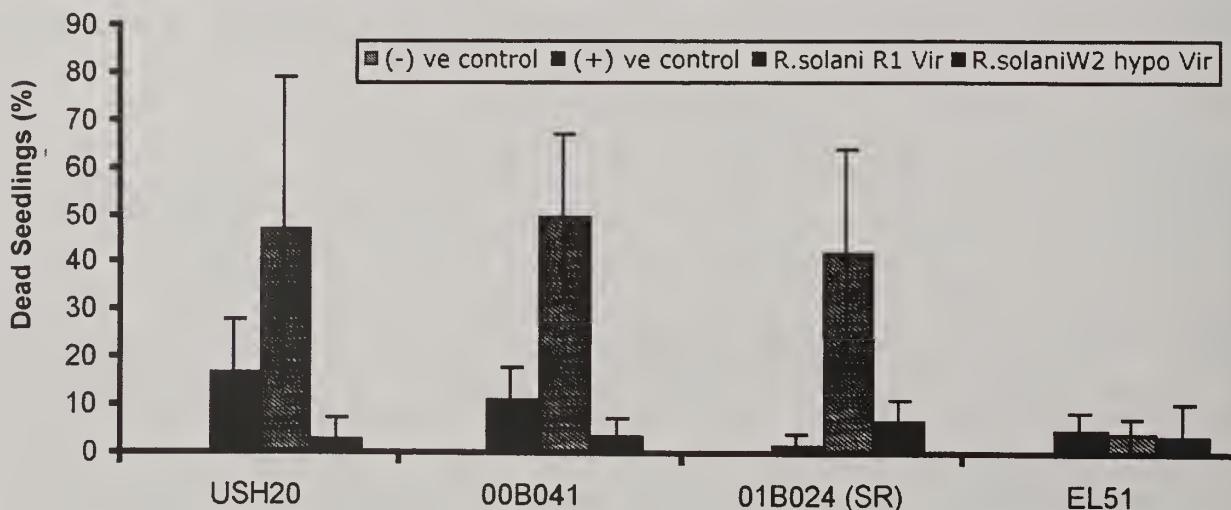
The field experiment was repeated in 2004 at the same location in an adjacent field plot from 2003. Only two entries (USH20 and EL51) were tested, using only AG2-2 R1 inoculant and sterile millet treatments. Eight replicate plots were used for each treatment. Inoculation was

done at the 4-leaf stage. All plots were super-inoculated at row closure with AG2-2 R1. Results in Table 1 indicate resistance to AG2-2 R1 is present in EL51, for both seedling and adult disease phases. USH20 showed a 10-fold reduction in survival between control and inoculated plots whereas EL51's stand was not significantly different with either treatment. Initial stand counts were similar between the entries (data not shown).

Table 1: Survival at harvest of inoculated and uninoculated plots of two varieties in 2004.

Treatment	Entry	Mean survival	std.dev.
Inoculated	USH20	0.87	0.83
	EL51	23.20	6.33
Control	USH20	8.87	4.44
	EL51	23.73	4.95

Changes in gene expression patterns may provide information about regulatory patterns and implicate potential biochemical pathways involved in expression of disease or resistance. There are no reports giving detailed assessment of the timing of deployment of plant genes following virulent and avirulent *R. solani* attack. The perception of the pathogen by the host as well as signal transduction activated by specific gene products is unknown for this specific host-pathogen system.

cDNA-AFLP was used to follow patterns of gene expression, and experiments are not complete. Preliminary analyses of fragments that were cut from cDNA-AFLP gels, cloned, and sequenced suggested that specific biochemical pathways might be deduced from this approach. Table 2 shows the similarities on nine such Transcript Derived Fragments (TDFs) with known proteins. The six TDFs from EL51 inoculated with AG2-2 (virulent) and analyzed 4 days after infection (PID4) could each have a role in limiting spread of Rhizoctonia in the tissue, however results need to be confirmed and extended to additional sequences.

Table 2: TDFs from control and inoculated sugar beet seedlings.

Treatment	BLAST detail	e value
Control USH20	Hypothetical protein (yeast)	3e-15
Control USH20	Xyloglucan endotransglucosylase/ hydrolase (Arabidopsis)	e-106
Control EL51	Hypothetical protein (yeast)	9e-11
EL51/Vir/PID4	Cystatin I precursor (Maize cysteine proteinase inhibitor)	1e-19
EL51/Vir/PID4	Mannose/glucose-specific lectin	3e-10
EL51/Vir/PID4	Drought Medicago truncatula cDNA	1e-36
EL51/Vir/PID4	Serine-rich protein (Yeast)	0.007
EL51/Vir/PID4	Jasmonate-induced protein	0.02
EL51/Vir/PID4	Glycine-rich RNA-binding protein	1e-30

Phenotypic and Genotypic Analyses of Root Sucrose Accumulation During Juvenile to Adult Developmental Phase Change in Sugar Beet

Daniele Trebbi and J. Mitchell McGrath

The objectives of this research were to understand the dynamic of root sucrose content during the early root developmental phases in sugar beet, to characterize gene expression profiles during these early developmental phases, and to identify genes differentially expressed between

developmental phases critical for sucrose accumulation. Greenhouse and field-grown plants were phenotypically analyzed and compared for two consecutive years, and gene expression profiles during root development were analyzed with cDNA-AFLP. Identification of genes differentially expressed between developmental phases characterized by different sucrose accumulation activity was performed by sequencing clones derived from a subtractive hybridization library. Quantification of the level of up and down-regulation of differentially expressed genes was performed with *Beta vulgaris* L. and *Arabdopsis thaliana* cDNA and oligonucleotide microarrays. Sucrose started accumulating after the 3rd week after emergence (WAE) and reached level comparable to mature root at the 7th WAE. Regulatory genes were mainly differentially expressed between these two stages of the root development in sugar beet.

Plants' life cycle can be subdivided in four chronologically successive phases: embryonic, juvenile vegetative, adult vegetative, and reproductive phases (Poethig, 1990). There are generally not specific physiological or morphological characteristics that indicate each specific phase, and phase changes are usually controlled by an array of signal transduction pathways triggered by the interaction of exogenous (i.e. temperature and light) and endogenous (i.e. hormones) factors. Environmental and genetic factors influencing the transition from adult vegetative to reproductive phases have been characterized in *Arabidopsis thaliana* as model organism, while phenotypic and genotypic characterizations during the transition from juvenile to adult vegetative phases have mainly been analyzed considering the shoot system of *Zea mays* (Poethig, 1990; Orkwiszewski and Poethig, 2000; Vega et al., 2002; Poethig 2003).

Little information is known on juvenile to adult vegetative phases transition in the plant root system. However, the timing and intensity of these biological changes, particularly related to sucrose accumulation during early root developmental of *Beta vulgaris* L., could influence the fast establishment of the crop, and could represent good indicators of productivity of lines in breeding programs and used as selection parameters. Theories on the dynamic of sucrose accumulation in sugar beet have been contradicting in early studies. Root development was believed to occur only late during the growing season after the complete development of the leaves apparatus, and sucrose accumulation trigger by lower temperatures at the end of the growing season (Bouillene *et al.*, 1940; Ulrich, 1952 and 1955). Watson and Selman (1938) and van Ginneken (1959) also assumed that early development was mainly dominated by leaves expansion, but were unable to distinguish separate phases of growth and sucrose accumulation in the root. Only during the 1970's the theory that photosynthates partitioning between leaves and root apparatus and sucrose accumulation in the root occur simultaneously and continuously during the growing season was accepted (Bergen, 1968; Follett et al., 1970; Milford, 1973, 1976, and 1988; Wise, 1979, 1980). Expression analysis of candidate genes involved in sucrose accumulation during root development in sugar beet has been addressed by several authors (Fieuw and Willenbrink, 1990; Chaubron *et al.*, 1995; Hesse *et al.*, 1995; Hesse and Willmitzer, 1996; Klotz and Finger, 2002; Klotz and Campbell, 2004). However, a comprehensive and comparative gene expression analysis during root development in sugar beet has rarely been investigated. Recently, Bellin *et al.* (2002) used a genome-wide comparative expression analysis between different sugar beet plant organs and were able to identify genes preferentially expressed in the storage root.

The objectives of this research were (i) to understand the dynamic of root sucrose content in greenhouse and field grown sugar beet lines during the early developmental phases in order to detect critical stages for sucrose accumulation; (ii) to characterize gene expression profiles

during the early developmental phases and to relate the dynamic of sucrose content with gene expression changes; (iii) to identify genes differentially expressed between developmental phases critical for sucrose accumulation.

A phenotypic analysis of greenhouse and field grown sugar beet lines was performed during the early root developmental phases to understand the dynamic of root sucrose content and to identify critical stages for sucrose accumulation. A genome-wide gene expression analysis (cDNA-AFLP) was also simultaneously performed to characterize gene expression profiles during these early developmental phases to correlate the dynamic of sucrose content with gene expression changes. Identification of genes differentially expressed between developmental phases critical for sucrose accumulation was finally performed using *Beta vulgaris* cDNA microarray selected by subtractive hybridization of a sugar beet root developmental cDNA library and *Arabidopsis thaliana* cDNA and oligonucleotides microarrays. These findings could identify critical root developmental stages for sucrose accumulation in greenhouse-grown plants that could be used as selective indicator during breeding programs; would detect the overall gene expression changes during root development indicating possible timing of shift from juvenile to adult vegetative phases, and relating this changes with the phenotypic changes observed at the root system level; and would finally identify genes mainly responsible for these changes, increasing our understanding on which metabolic pathways generally regulate the early stages of root development, and sucrose accumulation in particular, in sugar beet.

Materials and Methods

Plant material and growth conditions

Phenotypic analysis was investigated in greenhouse-grown plants during 2002 and 2003, and on field-grown plants during 2003. During 2002, 5 sugar beet lines characterized by different sucrose contents at root maturity (USH20, SR87, SR95, SR97, and SR96 ranging from 15 % to more than 17 % of sucrose on fresh weight, respectively), were grown in greenhouse for a period of 9 weeks after emergence (WAE). Experimental design was a randomized complete block design with three replications. In order to synchronize the time of emergence, seeds were treated with a 0.3 % H_2O_2 aqueous solution for 48 h (McGrath *et al.*, 2000), and germinated seeds showing root tip erupting the seed were hand-transplanted in 0.25 m^2 wooden boxes with 15 cm soil depth, spacing the seeds at 5 cm within and between rows, with an 8-16 dark-light cycle, at 15-20 °C, irrigated daily and fertilized twice a month. Time of emergence (March 4th, 2002) was estimated as 96 h after transplanting, when approximately 95 % of the plants were emerged.

Similarly, during 2003 two of the previously tested beet lines (USH20 and SR96) and the two parental lines of the mapping population, the sugar beet line C869 (Lewellen, 2004) and the table beet line W357B (Goldman, 1996), were grown in greenhouse for a period of 7 WAE. Experimental design was a randomized complete block design with two biological replications per each of the three replications. Time of emergence for the 2003 experiment was March 26th.

The same four beet lines tested in greenhouse during 2003 (USH20, SR96, C869, and W357B) were also analyzed for the dynamic of root sucrose accumulation in plants grown under field conditions during summer 2003. Each Line was planted in 7 consecutive double-rows (6 m long, with 0.76 m row inter-space) on May 21st, 2003, and time of emergence (June 2nd, 2003) was estimated when approximately 50 % of the plants were emerged. Plants were grown at Michigan State University Agronomy Farm, East Lansing, MI, using standard agronomic practices.

Samples collection and traits analysis

Plant samples were collected weekly from the 2nd to the 9th and from the 3rd to the 7th WAE, respectively for the 2002 and 2003 greenhouse experiments, and from the 3rd to the 20th WAE for the 2003 field experiment. Traits measured were root dry matter content and sucrose content on the root dry matter basis, while sucrose content on the fresh weight basis (% SucFW) was estimated from root dry matter content and sucrose content on the root dry matter basis. Root samples were dried by lyophilization, and root dry matter content (% DM) was estimated by comparison of weights before (w_{before}) and after (w_{after}) drying the samples, using the following equation:

$$\% \ DM = (w_{after} \ / \ w_{before}) \times 100$$

Sucrose content on the dry matter (% SucDM) was measured via HPLC for the 2002 and via enzymatic-fluorometric assay for the 2003 samples (Trebbi and McGrath, 2004), while sucrose content on the fresh weight (% SucFW) was estimated using the following equation:

$$\% \ SucFW = (\% \ DM \times \% \ SucDM) \ / \ 100$$

Plant size influenced the number of plants collected per sample (5 to 20 g). During the early weeks after emergence 50 to 60 plants per each sample were collected, and progressively reduced to 5 plants per samples during the final weeks. Plants collection for the greenhouse experiments was carried out to in order to leave the remaining plants thinned and equally spaced within each box, while plants collection in the field experiment was performed randomly selecting representative roots from the 7 double-rows plots and leaving the remaining plants equally spaced within plots. Phenotypic data of % DM, % SucDM and % SucFW of 2002 and 2003 experiments were analyzed and plotted with the non-linear regression model (logistic curve) using SigmaPlot 2001 software (SPSS Inc., Chicago, IL).

Analyses of the variation of gene expression (cDNA-AFLP experiments) and identification of differentially expressed genes during early roots developmental stages of sugar beet (microarrays experiments) were performed on greenhouse-grown plants during 2002 and 2003, simultaneously to the phenotypic analysis. Root samples for RNA extraction were collected weekly from the 1st to the 9th WAE from USH20 line during 2002, and from the 3rd to the 7th WAE from USH20 and SR96 lines during 2003. Plants were harvested and 5 g of roots were washed from soil, separated from leaves and hypocotyls, immediately frozen in liquid nitrogen and stored at $-$ 80 °C before RNA extraction. Two biological replications were collected per each line during 2003.

Nucleic acid purification

Total RNA was extracted using the Concert™ Plant RNA Reagent Kit (Cat. No. 12322-012, Invitrogen, Carlsbad, CA) and purified with RNeasy® Kit (Cat. No. 74104, Qiagen, Valencia, CA) as manufacturers instructions. Purified total RNA was then quantified using the RiboGreen® RNA Quantification Kit (Cat. No. R-11490, Molecular Probes, Eugene, OR) and RNA quality was assessed on Reliant® pre-cast 1.25 % agarose gels (Cat. No. R-54948, Cambrex, East Rutherford, NJ) as manufacturers instructions. Messenger RNA (mRNA) isolation from total RNA was obtained using the mTRAP™ Total Kit (Cat. No. 23012, Active Motif, Carlsbad, CA) performing the optional DNase treatment with DNaseI Amplification Grade (Cat. No. 18068-015, Invitrogen, Carlsbad, CA) as manufacturers instructions. Double-stranded complementary DNA (ds-cDNA) was synthesized using the SuperScript™ Double-

Standed cDNA Synthesis Kit (Cat. No. 11917-010, Invitrogen, Carlsbad, CA) employing the Oligo dT (Cat. No. N420-01, Invitrogen, Carlsbad, CA) as primer for first-strand cDNA synthesis, and quantification of the ds-cDNA was performed with the PicoGreen® DNA Quantification Kit (Cat. No. P-11496, Molecular Probes, Eugene, OR), following manufacturers instructions.

cDNA-AFLP protocol

For the sugar beet line USH20 samples collected in 2002 (1st to 9th WAE), cDNA-AFLP was performed using the *Eco*RI/*Mse*I (*E/M*) restriction enzyme pair combination and following the protocol of Bachem *et al.* (1996 and 1998) with modifications.

Complementary DNA (150 ng) was double digested at 37 °C for 3 hours with *Eco*RI (Invitrogen, Carslbad, CA) and *Mse*I (New England BioLabs, Beverly, MA) restriction enzymes in 30 $\Box$L of restriction solution [0.17 U $\Box$L^{-1} *Eco*RI; 0.17 U $\Box$L^{-1} *Mse*I; 10 mM Tris (pH = 7.5); 10 mM Mg Acetate; 50 mM K Acetate; 5 mM DTT; 0.05 $\Box$g $\Box$L^{-1} BSA]. Restriction enzymes were deactivated with a treatment at 70 °C for 15 minutes.

Double stranded AFLP adapters were produced from two single stranded non-phosphorilated linkers (MWG Biotech Inc., High Point, NC). To prepare non-phosphorilated double stranded *Eco*RI and *Mse*I adapters, equimolar amounts of the two complementary linkers, were slowly cooled down from 70 °C to 25 °C in a one-hour period, after an initial treatment at 95 °C for 3 minutes. *Eco*RI and *Mse*I adapters were ligated in 40 $\Box$L of ligation solution [0.35 $\Box$M *Eco*RI adaptor; 1.75 $\Box$M *Mse*I adaptor; 1 mM ATP; 0.025U $\Box$L^{-1} T4 ligase; 10 mM Tris (pH = 7.5); 10 mM Mg-Acetate; 50 mM K-Acetate] at room temperature overnight. The first PCR cycle of amplifications (pre-amplification) was performed with *Eco*RI (*E*+0) and *Mse*I (*M*+0) primers without any selective nucleotide extending into the restricted-legated genomic sequences. Two $\Box$L of restricted-ligated solution was used to prepare the 20 $\Box$L pre-amplification solution [0.5 $\Box$M *E*+0 primer; 0.75 $\Box$M *M*+0 primer; 0.188 mM dNTPs; 2.5 mM MgCl$_2$; 2 mM Tris-Hcl; 10 mM KCl; 0.01 mM EDTA; 0.1 mM DDT; 0.025 U $\Box$L^{-1} *Taq* polymerase], which was amplified for 20 cycles of 94°C denaturation (30s), 56°C annealing (30s) and 72°C extension (60s), with initial steps at 72°C for two minutes and 94°C for 1 minute, and last extension period at 72°C for 10 minutes. Pre-amplified solutions were five-fold diluted with low-TE buffer [10 mM Tris; 0.1 EDTA; pH = 8.0].

The second PCR cycle of amplifications (selective amplification) was performed using simultaneously (multiplexing) one of each IRD700 and IRD800 fluorescence-labeled (LI-COR Biosciences, Lincoln, NE) *Eco*RI primers with each three selective nucleotides (^{IRD}E+3), a single unlabeled *Mse*I primer with two selective nucleotides (*M*+2), and 1 $\Box$L of the diluted PCR product from the pre-amplification. Fifteen uL of selective amplification solution [17 nM $^{IRD700}E$+3 primer; 17 nM $^{IRD800}E$+3-primer; 0.5 $\Box$M *M*+2 primer; 0.188 mM dNTPs; 2.5 mM MgCl$_2$; 2 mM Tris-Hcl; 10 mM KCl; 0.01 mM EDTA; 0.1 mM DDT; 0.025 U $\Box$L^{-1} *Taq* polymerase] were amplified for 13 cycles of 94 °C denaturation (30 s), 65 °C annealing (30 s; -0.7 °C cycle^{-1}), and 72 °C extension (90 s), followed by 23 more cycles of 94 °C denaturation (30 s), 56 °C annealing (30 s) and 72 °C extension (90 s; +2 s cycle^{-1}), with initial denaturation at 94 °C for 30 s, and last extension period at 72 °C for 10 minutes. A total of 134 different primer combinations were analyzed combining 9 *E*+3 with all 16 possible *M*+2 primers (table available from the authors).

For the USH20 and SR96 samples collected in 2003 (from the 3rd to the 7th WAE; two

biological replications per sample), cDNA-AFLP was performed using the AFLP® Expression Analysis Kit (Cat. No. 830-06518, LI-COR Biosciences, Lincoln, NE) as manufacturer instructions, starting from 150 ng of ds-cDNA and using the *Taq*I/*Mse*I (*T/M*) restriction enzyme pair combination. Pre-amplified solutions were ten-fold diluted with low-TE buffer solution [10 mM Tris; 0.1 EDTA; pH = 8.0]. A total of 8 different primer combinations were analyzed combining 5 *T*+2 with 5 *M*+2 primers.

Transcript-derived fragments (TDFs) were separated via gel electophoresis. Two uL of stop/loading buffer [formamide 95% v/v; 10mM EDTA; 0.1% basic fuchsin; 0.01% bromophenol blue; pH = 9.0] were added to 5 uL of selective amplification product, and the solution was denatured at 96 °C for 5 minutes before gel separation. One uL of the denatured solution war analyzed on 4200 LI-COR IR^2 automated DNA sequencer (LI-COR, Biosciences, Lincoln, NE) in a denaturing 25 cm, 0.2 mm thick, 7 % acrylamide gel [7 M urea, 1x TBE (89 mM Tris-base; 89 mM boric acid; 2 mM EDTA), 3 mM ammonium per-sulfate, 4.4 mM TEMED] run at 1,500 V, at 45 °C for 4 hours. Gel images were collected, saved as image files and TDFs were scored to obtain binary datasets of polymorphic expression (band presence/absence) with the Saga^{MX} AFLP® Analysis Software (LI-COR, Biosciences, Lincoln, NE). Hierarchical cluster analysis using Pearson correlation similarity matrix was performed with Cluster 3.0 software and data visualized with Java TreeView 1.0.8 (Eisen *et al.*, 1998).

Sugar beet root development cDNA Library

In order to analyze gene expression during early root developmental stages a Uni-ZAP XR Vector cDNA library was commissioned to Amplicon Express, Pullman, WA. The library was obtained from 5 mg of total RNA derived from USH20 (4.5 mg) and SR96 (0.5 mg) lines grown during 2003, each representing equal amounts of 3rd, 4th, 5th, 6th, and 7th WAE samples.

Enrichment of differentially expressed genes

In order to enrich for differentially regulated transcripts between samples a Suppression Subtractive Hybridization (SSH) technique was employed (Diatchenko *et al.*, 1996) using the PCR-Select cDNA Subtraction Kit (Cat. No. K1804-1, DB Biosciences Clontech, Palo Alto, CA) as manufacture recommendations. Two different RNA samples were used for the SSH: a 3rd WAE sample obtained pooling equal amounts of total RNA from USH20 and SR96 lines grown in 2003, and a 7th WAE sample obtained pooling equal amounts of total RNA from USH20 and SR96 lines grown in 2003. Two different and independent transcript enrichments were performed: one that only enriched transcripts expressed at the 3rd but not at 7th WAE (early root development enriched transcripts), and the other that only enriched transcripts expressed at the 7th but not at 3rd WAE (late root development enriched transcripts). Specifically, to obtain early root development (3rd WAE) enriched transcripts, 2 □g of mRNA of each 3rd and 7th WAE were used for ds-cDNA synthesis. Both 3rd and 7th WAE cDNA samples were *Rsa*I digested, but adaptors ligation was only performed on the 3rd WAE digested cDNA (tester cDNA) but not on the 7th WAE digested cDNA (driver cDNA). Tester cDNA was subdivided in two pools and each pool was ligated to a different adaptor. A first hybridization was then performed using an excess of driver cDNA (10-fold higher) to each pool of tester cDNA heat denaturing the samples and allowing them to anneal. Most of the transcripts common to both samples formed ds-cDNA, while transcripts only present in the tester samples mainly remained as ss-cDNA, available for the second hybridization performed mixing together the two samples derived from the first hybridization with new denatured excess of driver cDNA. After the second hybridization new

differentially expressed ds-DNA hybrid with different adaptors at each ends were formed in solution, which were selectively amplified using adaptors-specific primers with two consecutive PCR amplifications. Similarly, to obtain late root development (7th WAE) enriched transcripts, the same procedure was performed using the 7th WAE sample as tester cDNA and the 3rd WAE sample as driver cDNA. Samples were phenol: chloroform: isoamyl alcohol (25:24:1) purified, ethanol precipitated and resuspended in 30 uL sterile RNase- and DNase-free water. Early and late root development enriched transcripts were used as probes for cDNA library hybridization.

Library hybridization and cDNA sequencing

The cDNA primary library obtained from Amplicon Express was titered to estimate the original concentration of plaques forming units (pfu) following the Uni-ZAP® XR Premade Library Protocol (Stratagene, La Jolla, CA). *E.coli* host bacteria XL1-Blue MRF' strain was grown on LB-tetracycline agar plate [NaCl 10 g L^{-1}; tryptone 10 g L^{-1}; yeast extract 5 g L^{-1}; agar 20 g L^{-1}; pH 7.0; tetracycline 15 mg L^{-1}] at 37 °C overnight, and 50 mL of LB broth with supplement [NaCl 10 g L^{-1}; tryptone 10 g L^{-1}; yeast extract 5 g L^{-1}; pH 7.0; 10 mM $MgSO_4$; 0.2 % (w v^{-1}) maltose] were inoculated with a single colony and incubated at 37 °C for 6 h. Cells were precipitated and resuspended in 10 mM $MgSO_4$ solution at a concentration equal to an optical density at 600 nm (OD_{600}) of 0.5. Four ten-fold dilutions of the primary library were incubated with 200 □L of XL1-Blue MRF' cells at OD_{600} = 0.5 for 15 min at 37 °C and plated on NZY agar [NaCl 5 g L^{-1}; $MgSO_4$ - 7 H_2O 2 g L^{-1}; yeast extract 5 g L^{-1}; NZ amine 10 g L^{-1}; agar 15 g L^{-1}; pH 7.5] plates with 15 □L 0.5 M IPTG and 50 □L X-gal (250 mg mL^{-1}). Bacteria were grown overnight at 37 °C and white and blue plaques were counted. Ten uL of primary library, representing an estimate of 25,500 pfu, were plated with XL1-Blue MRF' strain cells on ten 150 mm diameter NZY agar Petri dishes (Cat. No. 25384-326, VWR, Bristol, CT) and grown overnight, obtaining an average density of ~15 pfu cm^{-1}. Plaque lifts was performed with Nytran® 0.2 □m nylon membranes (Cat. No. 78112, Schleicher & Schuell, Keene, NH) placing a dry membrane onto the NZY agar for 5 min. A denaturing step was performed placing the membrane face-up on 3MM whatmat filter papers (Whatman International, Kent, England) pre-soaked in 1.5 M NaCl and 0.5 M NaOH solution for 5 min. A neutralizing step was performed transferring the membrane face-up on 3MM Whatman papers pre-soaked in 1.5 M NaCl and 0.5 M Tris (pH 8.0) solution for 5 min, followed by a final rising step where the membrane was transfer face-up on 3MM Whatman paper pre-soaked in 0.1 M Tris-HCl (pH 7.5) and 2× SSC solution for an additional 5 min. Two consecutive plaque lifts were performed per each plate. Nucleic acids were UV cross-linked (0.12 J cm^{-2} for 30 s) to the membranes. Two plaque lifts were performed per each Petri dish. In order to facilitate plaque transfer to the membrane during the second lift, membranes were pre-wetted the a 5× SSC solution, dried on 3MM Whatman papers for 3 min and plaque transfer from agar was allowed for 10 min. NZY agar plates were stored at 4 C for post-hybridization plaques collection.

Early and late root development enriched transcripts obtained from SSH technique were ^{32}P labeled and used as probes for membranes hybridization. Amplified transcripts (100 ng) were denatured at 100 °C for 5 min and ^{32}P was incorporated in the 50 □L labeling solution [^{32}PdCTP 50 □Ci; dATP, dTTp and dGTP 0.6 mM; Tris 1.5 mM; $MgCl$ 0.15 mM; random hexamer; 2-mercaptoethanol; BSA 0.4 ng □L^{-1}; Hepes; Klenow enzyme 0.05 U □L^{-1}] after overnight incubation at room temperature. Two separate reactions were performed for each of the 3rd and 7th WAE enriched transcripts. Unincorporated ^{32}PdCTP was removed from the samples by filtration and pre-hybridization was performed placing the membranes (20) in 2 L of pre-

hybridization solution [6× SSPE; 6× Denhardt's solution; 0.5 % SDS; 100 □g mL^{-1} denatured fish sperm] at 60 °C for 20 h.

Membranes hybridization was performed separately for early and late labeled enriched transcripts. The 10 membranes derived from the first plaque lifts were hybridized with late enriched transcripts (7th WAE) in 100 mL pre-hybridization solution, at which 3 mL labeled probe were added, and incubated at 60 °C for 16 h. Similarly, the 10 membranes derived from the second plaque lifts were hybridized with early enriched transcripts (3rd WAE). Post-hybridization membranes washes were performed in 2× SSC and 1 % SDS solution at room temperature for 5 min, followed by a second wash in 2× SSC and 1 % SDS solution at 65 °C for 15 min, and a final wash in 2× SSC and 0.1 % SDS solution at 65 °C for 15 min. Membranes were exposed to X-ray films (Hyperfilm™ MP, Cat No. RPN30K, Amrsham Pharmacia, Piscataway, NJ) for 7 days before film developing. The number and relative positions in the NZY agar Petri dishes of plaques that hybridized to the probes were analyzed. Plaques were classified based on which kind of probe they hybridized with. Three classes were observed: 3rd WAE plaques, 7th WAE plaques, and 3rd & 7th WAE plaques, which respectively hybridized only to the 3rd WAE probe, only to the 7th WAE probe, or to both probes. Plaques were collected from NZY agar with sterile Pasteur pipette in 500 □L SM buffer [NaCl 5.8 g L^{-1}; MgSO$_4$ * 7 H$_2$O 2 g L^{-1}; 50 mM Tris-HCl pH 7.5; 0.01 % w v^{-1} gelatin] and 25 □L chloroform. Single clone excision of the pBluscript phagemid from the Uni-ZAP XR vector was performed using the ExAssist helper phage and the *E.coli* host bacteria SOLR strain, following the Uni-ZAP$^{®}$ XR Premade Library Protocol. pBluscript-containing phages eluted from the collected plaques were incubated with *E.coli* host bacteria XL1-Blue MRF' strain and ExAssist helper phage at 37 °C overnight. Cell suspension was heated at 70 °C for 20 min, centrifuged at 1000g for 15 min, and the supernatant containing the excised pBluscript was used to inoculate *E.coli* host bacteria SOLR strain on LB-ampicillin agar plates at 37 °C overnight. Single colonies were transfer to LB freezing media [NaCl 10 g L^{-1}; tryptone 10 g L^{-1}; yeast extract 5 g L^{-1}; pH 7.5; 4 % glycerol; K$_2$HPO$_4$ 36.2 mM; KH$_2$PO$_4$ 13.2 mM; sodium citrate 1.7 mM; MgSO$_4$ 4 mM; ammonium sulfate 6.8 mM; ampicillin 100 mg L^{-1}] and sent to the Genomic Support Technology Facility at Michigan State University for clone sequencing. Sequences were submitted to GenBank (http://www.ncbi.nlm.nih.gov/) and blasted (Altschul et al., 1990).

Beta vulgaris cDNA microarray

Sequenced cDNA clones selected with enriched transcripts were also spotted on microarrays and hybridized with original, non-enriched total RNA from early and late root developmental stages in order to verify their temporal variation in expression and quantify their relative abundance. RNA samples from two different biological replications of each 3rd WAE and 7th WAE samples of both USH20 and SR96 lines grown during 2003 were used as probes.

To spot cDNA clones on microarrays, 2 □L of the plasmid solutions used by the Genomic Support Technology Facility at Michigan State University for clones sequencing were used to PCR amplify cDNA inserts using T7 (5'-TAA TAC GAC TCA CTA TAG GG-3') and T3 (5'-ATT AAC CCT CAC TAA AGG GA-3') primers for 30 cycles of 94°C denaturation (20s), 52°C annealing (20s) and 72°C extension (90s), with initial steps at 94°C for two minutes and last extension period at 72°C for 5 minutes. Amplified DNA was precipitated in 200 □L of precipitation solution [150 mM Na acetate; 90 % ethanol] at − 80 °C for 2 h, centrifuged at 5000 *g* for 1 h, precipitate was washed with 200 □L 80 % ethanol solution, centrifuged at 5000 *g* for

30 min, and air-dried overnight. Pellet was resuspended in 25 $\Box$L of slide-printing solution (3×
SSC) and 0.5 $\Box$L run on 1 % agarose gel to verify a single insert was amplified from each clone.

In addition to the *B. vulgaris* cDNA selected from the library via subtractive probes, 17 more
clones, representing different classes of polyubiquitins, ribosomal proteins, tubulin, GAPDH,
and actins, that did not show any changes in gene expression between the 3rd and 7th WAE RNA
samples used in the *A. thaliana* oligonucleotide microarray, were selected as controls for spot
intensity normalization during data analysis.

Amplified clones were submitted to the Genomic Support Technology Facility at Michigan
State University and printed in triplicate on each slide array. A total of 8 microarrays were
overall analyzed, two for comparing the 3rd WAE and 7th WAE samples from the first biological
replications of the USH20 line; two for the 3rd WAE and 7th WAE samples from the second
biological replications of the USH20 line; two for the 3rd WAE and 7th WAE samples from the
first biological replications of the SR96; and two for the 3rd WAE and 7th WAE samples from the
second biological replications of the SR96 line. For each one of these four comparisons two
technical replications were performed inverting the cyanine dyes between samples (i.e. 3rd
WAE–Cy3 labeled and 7th WAE–Cy5 labeled for the first slide, and 3rd WAE–Cy5 labeled with
7th WAE–Cy3 labeled for the second slide). RNA samples preparation and microarray
hybridization were performed as described in c, with the exception of the hybridization
temperature that was set at 54 °C for 16 h.

Normalization of clone signal intensities between microarrays was calculated using average
intensities of the 17 controls. Slide hybridization was detected with Affymetrix 428 Array
Scanner (Affymetrix, Santa Clara, CA), and data were analyzed with GenePix Pro 3.0 (Axon
Instruments, Union City, CA), and normalized using the Statistics for Microarray Analysis
Software 0.5 (http://www.stat.berkeley.edu) and the R Statistical Package
(http://lib.stat.cmu.edu/R/CRAN) (Yang et al., 2002). In addition to the software default
thresholds parameters used to define presence versus absence of hybridization on each printed
spot, three more thresholds parameters were employed to eliminate doubtful hybridizations: (i)
both technical replications were discarded from further analysis if in at least one replication both
Cy3 and Cy5 mean signal intensities were lower than 1000 units, or (ii) if they were lower than
the double of the Cy3 and Cy5 background median signal intensities, or (iii) if the area of both
Cy3 and Cy5 with mean signal intensities lower than background median signal intensities plus 1
background standard deviation were higher than 50 % of the spot surfaces. For those spots with
positive hybridization, fold-level differences of gene expression between early and late root
development were estimated from the ratios of the intensities of the dyes signals per each gene.
These differences were calculated as the ratio between the Cy5 hybridization mean intensity
($Cy5_{Hm}$) minus the Cy5 background median intensity ($Cy5_{Bmed}$) and the Cy3 hybridization mean
intensity ($Cy3_{Hm}$) minus the Cy3 background median intensity ($Cy3_{Bmed}$) as follows:

$$(Cy5_{Hm} - Cy5_{Bmed}) / (Cy3_{Hm} - Cy3_{Bmed}) \qquad \text{if ratio was} \geq 1$$

or as follows:

$$-1 / [(Cy5_{Hm} - Cy5_{Bmed}) / (Cy3_{Hm} - Cy3_{Bmed})] \qquad \text{if ratio was} < 1$$

Individual clone average signal hybridization intensity and standard deviation for confidence
intervals estimation were calculated from the 6 replications (3 spots per each of the 2 slides) of
the same biological replication comparison. Clones were considered present if hybridization was

observed in at least one of the four biological replications (two per each sugar beet line), with at least two of the six replicated spots with signal hybridization intensity higher than the threshold values.

Within each of the four biological replications, genes were considered differentially expressed if the lower CI_{90} was ≥ 1.1-fold expression difference between the two developmental stages (up-regulated genes during late root development), or if the higher CI_{90} was ≤ -1.1-fold expression difference (down-regulated genes during late root development). Experimental-wise, genes were considered differentially expressed if they showed at least two of the four biological replications with significant differences in the level of gene expression. If a gene showed one biological replication differentially expressed in one direction, it was considered as variable (V) if another replication did show variation of gene expression in the opposite direction, or as no change (NC) if none of the other biological replications showed significant signal hybridizations.

To summarize line-specific and overall variation of gene expression two estimate of differential gene expression (DGE) were performed. For each clone, differential gene expression was estimated independently for both USH20 and SR96 sugar beet lines (DGE_{line}) averaging the level of expression of the two biological replications, as follows:

$$DGE_{line} = [(r_{t1} \times DGE_{t1}) + (r_{t2} \times DGE_{t2})] / (r_{t1} + r_{t2})$$

where r_{t1} and r_{t2} are the number of technical replications of the first and second biological replications of a specific line, and DGE_{t1} and DGE_{t2} are the samples fold difference of gene expression of the first and second biological replications of the same line. Also, for each clone a mean differential gene expression (DGE_m) was estimated combining data from the two sugar beet line as follows:

$$DGE_m = [(r_{1line1} \times DGE_{line1}) + (r_{1line2} \times DGE_{line2})] / (r_{line1} + r_{line2})$$

where r_{line1} and r_{line2} are the total number of technical replications in both biological replications of the first and the second beet lines respectively, and the DGE_{line1} and DGE_{line2} are the differential gene expression of the first and the second beet lines.

Functional classification of differentially expressed genes was performed using the Munich Information Center for Protein Sequences (http://mips.gtf.de).

Arabidopsis thaliana cDNA microarray

Complementary DNA Microarrays carrying 12,580 PCR-amplified and purified cDNA derived from *Arabidopsis thaliana* developing seeds (White et al., 2000) were provided by the Genomic Support Technology Facility at Michigan State University. Of these 12,580 cDNA, 5791 identified transcripts with known or putative functions, 2322 with proteins with unknown functions, 3572 with hypothetical proteins, and 895 had no sequence similarity to other known nucleotide sequences in public databases.

Sugar beet total RNA samples from the 3rd and 7th WAE of the USH20 line grown during 2002 was used to hybridize the *A. thaliana* cDNA microarrays following the TIGR Aminoallyl Labeling of RNA for Microarray Protocol (http://www.tigr.org). Specifically, 20 ug of total RNA were combined with 6 ug of oligo dT [an equimolar mixture of individually synthetized $T(T)_nTA$, $T(T)_nTC$, and $T(T)_nTG$] in a final 18.5 uL volume and incubated at 70 °C for 10 min. First strand cDNA synthesis with Uracil-aminoallyl incorporation was performed overnight at 42 °C, using the SuperScript II RT Kit (Cat. No. 18064-014, Invitrogen life technologies, Carlsbad,

CA) and Aminoallyl-dUTP (Cat. No. A-0410, Sigma-Aldrich Corp., St. Louis, MO) in a 30 uL reaction solution following manufactures instructions. RNA strands were hydrolyzed with 10 uL of 1 M NaOH and 10 □L of 0.5 M EDTA and after incubation at 70 °C for 15 min. Unincorporated d-UTP was removed with the QIAquick PCR Purification Kit (Cat. No. 28106, Qiagen, Valencia, CA), replacing PE and EB buffers from Qiagen Kit with Phosphate Wash Buffer [5 mM KPO_4, pH 8.5, 80% ethanol] and Phosphate Elution Buffer [4 mM KPO_4, pH 8.5], respectively. Aminoallyl-cDNA was coupled with cyanine dyes Cy3 ester and Cy5 ester (respectively Cat. No. PA23001 and PA25001, Amersham Pharmacia, Piscataway, NJ) in 9 uL coupling solution [0.1 M Na_2CO_3, pH 9] for 1 h at room temperature and protected from light. A second purification with the QIAquick PCR Purification Kit was performed to remove unlinked dyes as manufacture instructions, and labeled single strand cDNA was resuspended in 4 □L of 10 mM EDTA for slide hybridization. Third and 7th WAE samples were both individually labeled with each dye and two technical replications were performed inverting the dyes between samples during slides hybridization; specifically, 3 WAE–Cy3 labeled and 7 WAE–Cy5 labeled were used for the first slide, and 3 WAE–Cy5 labeled with 7 WAE–Cy3 labeled were used for the second slide. Pre-hybridization treatments of the microarray slides were performed with two consecutive washes in 0.1 % SDS in a Coplin jar for two min each, followed by two more washes in deionized filtered water for two min and final treatment in 100 °C water for 3 min. Slides were then re-washed in deionized filtered water at room temperature and dried by centrifugation at ~ 200 g for 5 min. Slide hybridization was performed with SlideHyb™ Buffer (Cat. No. 8861, Ambion, Austin, TX) following manufacture recommendations. The 4 □L of resuspended labeled cDNA were denatured at 96 °C for 10 minutes and mixed with 70 □L of pre-warmed (65 °C) SlideHyb™ Buffer. Solutions were loaded in the microarrays and hybridization was performed at 48 °C for 16 h. Three 15 min post-hybridization washing treatments at increasing stringency were performed after hybridization with low [2× SSC, 0.5 % SDS], medium [0.1× SSC, 0.2 % SDS], and high [0.1× SSC] stringency solutions. Data analysis was performed with GenePix Pro 3.0 software (Axon Instrument, Union City, CA) with the same threshold parameters used in the *B. vulgaris* cDNA microarray experiment. Folds expression differences at both 3rd and 7th WAE were averaged between technical replications and 90 % confidence intervals (CI_{90}) for the mean fold difference was estimated with the two-sided t-test. Genes were considered differentially expressed if the lower CI_{90} was ≥ 1.1-fold expression difference between the two developmental stages (up-regulated genes during late root development), or if the higher CI_{90} was ≤ -1.1-fold expression difference (down-regulated genes during late root development).

In order to comparatively estimate the quantity of transcripts from the hybridization intensity signals (on a scale from 0 to 65,000 signal units), clones were subdivided as having low, medium, high, or very high hybridization signal, if the average hybridization mean intensities of the dyes of both replications were < than 2000, between 2000 and 4000, between 4000 and 10,000, or > 10,000 signal units, respectively. Functional classification of differentially expressed genes was performed using the Munich Information Center for Protein Sequences (http://mips.gtf.de).

Arabidopsis thaliana oligonucleotide microarray

Sugar beet total RNA samples from 3rd WAE and from 7th WAE of the USH20 line grown during 2003 were used to hybridize *A. thaliana* oligonucleotide microarrays following the Affimetrix GeneChip® Expression Analysis Protocol. Specifically, 16 ug of purified total RNA

were used for ds-cDNA synthesis using the SuperScript II Double Stranded cDNA Synthesis Kit (Cat. No. 11917-010, Invitrogen life technologies, Carlsbad, CA) and the T7-Oligo(dT) Promoter Primer Kit (Cat. No. 900375, Affymetrix, Santa Clara, CA) as manufactures recommendations. Complementary DNA was purified adding an equal volume (162 $\Box$L) of phenol: chloroform: isoamyl alcohol (25:24:1) (Cat. No. 15593-031, Invitrogen, Carlsbad, CA) and separating the phases in Phase Lock Gel Heavy 2 mL (Cat. No. 955154045, Eppendorf, Hamburg, Germany). Complementary DNA was ethanol precipitated adding 0.5 volume (81 uL) of 7.5 M ammonium acetate, 3.5 volumes (567 uL) of ice-cold absolute ethanol and 4 $\Box$L of 40× glycogen (Cat. No. 901393, Roche, Penzberg, Germany), and resuspended in 12 $\Box$L RNase-free water. Complementary RNA (cRNA) was synthesized from the purified ds-cDNA template with T7 RNA polymerase and biotin-labeled nucleotides using the BioArray™ HighYield™ RNA Transcript Labeling Kit (Cat. No. 42655-10, ENZO Life Sciences, Farmingdale, NY) as manufacture recommendations. Complementary RNA was purified with RNeasy® Kit (Cat. No. 74104, Qiagen, Valencia, CA), quantified with RiboGreen® RNA Quantification Kit (Cat. No. R-11490, Molecular Probes, Eugene, OR) and cRNA quality was controlled on Reliant® pre-cast 1.25 % agarose gels (Cat. No. R-54948, Cambrex, East Rutherford, NJ) as manufacturers instructions. Hybridization of the fragmented cRNA was performed at the Genomic Support Technology Facility at Michigan State University using the Arabidopsis Genome ATH1 Array (Cat. No. 900385, Affymetrix, Santa Clara, CA). Per each 3rd and 7th WAE cRNA sample a single array was used. Data analysis was performed using two different approaches. Positive hybridizations and changes in gene expression were derived (i) using the Affymetrix Suite software with default threshold parameters optimized for *A. thaliana* transcripts hybridization ($\Box_1 = 0.05$; $\Box_2 = 0.065$; $\Box_1 L = \Box_1 H = 0.0045$; $\Box_2 L = \Box_2 H = 0.006$; $\Box = 0.015$; d =1.1; TGT = 100), or (ii) using a non-conventional analysis based on the level and the ratio of probe set signals hybridization of the two microarrays to estimate presence and changes of expression between samples. For the non-conventional analysis, normalization of probe set signal intensities between arrays was calculated using average intensities of 64 internal controls. Probe set was considered present if signal hybridization was ≥ 400, and hybridization strength was estimated as low (L), medium (M), high (H) and very high (VH), if signal hybridization was between 400 to 500, between 500 to 600, between 600 to 1000, or > 1000 units, respectively. Functional classification of differentially expressed genes was performed using the Munich Information Center for Protein Sequences (http://mips.gtf.de).

Results

A phenotypic analysis of greenhouse and field grown sugar beet lines during the early developmental phases was performed to understand the dynamic of root sucrose content and to identify critical stages for sucrose accumulation. A genome-wide gene expression analysis (cDNA-AFLP) was performed simultaneously the phenotypic analysis to characterize gene expression profiles during the early developmental phases and to correlate the dynamic of sucrose content with gene expression changes. Identification of genes differentially expressed between developmental phases critical for sucrose accumulation was finally performed using *Beta vulgaris* cDNA microarray selected by subtractive hybridization of a sugar beet root developmental cDNA library, and *Arabidopsis thaliana* cDNA and oligonucleotides microarrays.

Phenotypic analysis

All the five sugar beet lines grown under greenhouse conditions for a period of 9 WAE in

2002 showed the same trend of sucrose accumulation expressed on the fresh weight basis. Sucrose mainly accumulated in roots from the 3rd to the 7th WAE, increasing from less than 1 % to more than 10 % SucFW during this period. Differences ($p < 0.05$) between lines were only observed at the 9th WAE between the higher sucrose accumulator line SR96 (13.5 % SucFW) and the lower sucrose accumulator line USH20 (10.7 % SucFW). This difference of root sucrose content on the fresh weight basis was mainly caused by differences of root dry matter content between lines more than differences in sucrose content on the dry matter basis. Root dry matter showed the same trend of sucrose accumulation on the fresh weight basis, and varied from an average of 7 % DM at the 2nd WAE to an average of 22 % DM at the 9th WAE, when also a difference ($p < 0.05$) between SR96 (23.3 % DM) and USH20 (19.5 % DM) was observed. Differently, sucrose accumulation expressed on the dry matter basis varied from an average of 5 % SucDM at the 2nd WAE to an average of 55 % SucDM at the 9th WAE, but no differences were observed between lines.

For the 2003 greenhouse experiment, only the two extreme SR96 and USH20 lines were analyzed, using two biological replications per line, and focusing on the period between the 3rd and the 7th WAE. A similar trend of sucrose accumulation on the fresh weight basis was observed in 2003 respect the previous year, with sucrose content increasing from 1 % at the 3rd WAE to more than 12 % SucFW at the 7th WAE. The level of root sucrose content reached at the 7th WAE in 2003 was similar to the level reached at the 9th WAE in 2002, but no difference was observed between lines. However, similarly to the results obtained in 2002, SR96 accumulated more root dry matter (23.8 % DM) respect USH20 (21.5 % DM) at the 7th WAE in 2003. The two parental lines of the mapping population were also analyzed in the 2003 greenhouse experiment to analyze phenotypic differences between sugar and table beets. The sugar beet line C869 had similar trends and levels of dry matter and sucrose accumulation, expressed on both the fresh and dry matter basis, respect the two other sugar beet lines, but showed a dry matter content lower (20.1 % DM) than the USH20 line. The table beet line W357B accumulated a significantly lower ($p < 0.001$) amount of sucrose on the fresh weight (8.4 % SucFW at the 7th WAE) respect the sugar beet lines, caused by a decrease of both root dry matter content (18.2 % DM at the 7th WAE) and sucrose on dry matter basis (46.4 % SucDM at the 7th WAE).

To evaluate levels of root dry matter and sucrose accumulation in field-grown plants and to compare these levels with the results obtained in greenhouse-grown condition, SR96, USH20, C869 and W357B lines were field-grown during 2003 and analyzed weekly from the 3rd to the 20th WAE. Root sucrose content on the fresh weight increased from 2 % at the 3rd WAE to 16/17 % SucFW at the 20th WAE in the sugar beet lines and to 9 % SucFW in the table beet line. No differences were observed between sugar beet lines, which showed higher ($p < 0.05$) sucrose content on the fresh weight respect the table beet line starting at the 6th WAE. Root dry matter increased from 12 % DM at the 3rd WAE to 23 % DM at the 20th WAE in the sugar beet lines and to 16 % DM in the table beet line (Fig. 4.8), with difference between sugar and table beet lines but no difference between sugar beet lines. Similarly, sucrose content on the dry matter basis increased from an averages of 18 % SucDM at the 3rd WAE to 70 % SucDM at the 20th WAE in the sugar beet lines and to 55 % SucDM in the table beet line (Fig. 4.9), with difference between sugar and table beet lines but no difference between sugar beet lines.

Overall, greenhouse-grown plants anticipated % DM and % SucFW traits values observed in field-grown plants. Values observed at the 9th WAE in 2002 and at the 7th WAE in 2003, were

usually detected between the 13th and the 16th WAE in field-grown plants for both % DM and % SucFW traits. Differently, values of sucrose content on the dry matter basis in greenhouse-grown plants were indicative of the same values observed in field-grown plants after similar growing periods.

Complementary DNA-AFLP

Differential gene expression analysis of root tissues sampled from the 1st to the 9th WAE during 2002 showed that the 134 different primer combinations yielded a total of 3302 transcript-derived fragments (TDFs) (24.6 TDFs per primer combination). A total of 2,181 (67 %) TDFs did not show any differential level of expression between samples (monomorphic TDFs), while 1121 (33%) showed difference (absence versus presence) of expression in at least two of the nine samples analyzed (polymorphic TDFs). Overall, the total number of TDFs per week decrease form an average of ~2900 in the first 3 weeks to an average of ~2700 from the 4th to the 9th WAE. Of the polymorphic fragments, 199 (6.0%) were only expressed in at least two samples of the first five WAE and never observed later (early-expressed TDFs), 109 (3.3 %) were only expressed in the last five WAE and never earlier (lately -expressed TDFs), while 50 (1.5 %) were only expressed between the 3rd and 7th WAE but never during the first or last two weeks of analysis. Cluster analysis based on similarity of gene expression profiles of the different periods of root development showed continuous changes from the 1st to the 5th WAE and from the 6th to the 9th WAE, with a marked shift in gene expression between the 5th and the 6th WAE. The 8 different primer combinations analyzed on the two biological replications of the two USH20 and SR96 lines between the 3rd and 7th WAE in 2003, overall confirmed the results observed in the 2002 also estimating the potential differences between lines and the reliability of the cDNA-AFLP technique. SR96 showed a higher level of amplified transcripts and polymorphism respect USH20. The 8 *TaqI/MseI* primer combinations used in 2003 yielded a total of 457 TDFs, of which 373 common between lines, 34 specific for USH20 and 50 specific for SR96. A total of 127 TDFs (27.8 %) were polymorphic, of which 80 common between lines, 11 specific for USH20 and 36 specific for SR96. On average, only 4.2 % of TDFs showed difference of expression between biological replications (presence and absence of fragment in the same replication, in contrast with both absence or both presence) for both lines. Clustering analysis confirmed the same trend of gene expression previously observed, with a shift of expression between the 3rd and the 7th WAE, corresponding with the phenotypic increase of root sucrose accumulation.

Library hybridization and cDNA sequencing

The cDNA library obtained from Amplicon Express showed an average insert size of 1.2 kb and an estimated titer of 2.55 x 10^6 pfu mL^{-1}, of which 2.25 x 10^6 pfu mL^{-1} recombinant. A total of 966 plaques hybridized to the SSH probes and were collected and their inserts sequenced. Of these, 587 hybridized only to early enriched transcripts (3rd WAE), 313 hybridized only to late enrich transcripts (7th WAE), while 66 hybridized to both probes. Sequences quality controls revealed that a total of 710 inserts were successfully sequenced, showing an average reading length of 769 (SD ±156) nucleotides, while the other clones mainly matched vector sequences or presented very short readings. These 710 sequences were deposited in GeneBank and represented 442 (GeneBank No. CV301334 to CV301775), 219 (GeneBank No. CV301776 to CV301994), and 49 (GeneBank No. CV301285 to CV301333) transcripts that hybridized only with early, only with late, or to both developmental stages enriched probes, respectively. To evaluate sequence efficiency some of the clones were sequenced in duplicate or triplicate, and 63

of the 710 sequences represented 29 individual sequences. Considering these 29 sequences plus the 647 sequences for which single-pass sequence was performed, clustering analysis revealed that of these 676 transcripts, 267 grouped in 84 clusters, ranging from 2 to 14 sequences per cluster. Sixty-two of these sequences grouped in 5 clusters representing transcripts that mainly hybridized with the 3rd WAE (glycine-rich RNA-binding protein, jacalin lectin family protein similar to agglutinin, elongation factor 1-alpha, and an unknown protein) or mainly with the 7th WAE (pentatricopeptide (PPR) repeat-containing protein) enriched probes. The majority of the clusters (50) were composed by transcripts that hybridized only to one but not to the other enriched probes, while the other clusters contained transcripts that hybridized both early and late enriched transcripts. Other than the 84 transcripts represented by the 267 clustered sequences, another 247, 137, and 25 individual (singletons) transcripts hybridized to the 3rd, 7th, or to both subtractive probes respectively, giving a total of 493 non-redundant individual transcript sequences.

Beta vulgaris cDNA microarray

The overall level of clone hybridization in the subtracted library-derived microarray showed that of the 676 *B. vulgaris* individual clones, 551 (81.5 %) were present in the original total RNA samples. The majority of the clones (366 = 54.1%) were present in all four biological replications, while only 21 clones (3.1 %) were only present in one biological replication and were not considered for differential gene expression analysis. No difference in the frequency of presence versus absence was observed between the 267 clustering sequences (217 = 81.3 %) and the 409 singleton sequences (334 = 81.7 %). However, a higher proportion of clustering sequences hybridized to all four biological replications (178/217 = 82.0 %) respect the singleton ones (188/334 = 56.3 %).

Of the 551 clones detected as present, 95 (17.2 %) and 110 (20.0%) were up- and down regulated respectively, while 21 (3.8 %) clones had variable pattern of gene expression between biological replications and 325 (59.0 %) did not show any significant variation of expression. Most of the differentially expressed clones hybridized to all 4 replications, 88 (92.6 %) 99 (90.0 %) and 20 (95.2 %) for the up-, down-, and variable-regulated respectively, while only 159 (48.9 %) of those clone that did not show any variation of expression hybridized to all replications.

Transcripts that hybridized with early and late enriched probes were expected to respectively show down- and up-regulation during the subtracted library microarray experiment. The comparison of the level of expression of differentially expressed genes with the classes of subtracted probe the clone hybridized with, indicated a good accuracy of the SSH technique, particularly on clones that hybridized to at least three biological replications. Specifically, of the 95 up-regulated clones 61 (48 + 13 = 64.2 %) hybridized to late root development enriched transcripts (7th WAE), while of the 110 down-regulated clones, 93 (87 + 6 = 84.5 %) hybridized to early root development enriched transcripts (3rd WAE), as expected. The proportions increase to 47/60 (78.3 %) for up-regulated and to 57/63 (90.5 %) for down-regulated genes if considering only clones that hybridized to at least three biological replications. Interestingly, 38 transcripts, 12 up-regulated and 26 down-regulated, are differentially expressed only in SR96 line, while only 2 transcripts (up-regulated) are specific of the USH20 line.

The 95 up-regulated clones represented a total of 55 non-redundant sequences, 23 derived from 62 originally clustered sequences, and 32 from singleton sequences. Of the clustered sequences, 12 sequences, which all hybridized with the 7th WAE enriched probes, were

represented by the pentatricopeptide (PPR) repeat-containing protein cluster, showing an average level of 2.6-fold over-expression during late root development. Similarly, all sequences of the dormancy/auxin-repressed family protein (5 sequences 6.9-fold up-regulated), of the nucleoporin family protein (4 sequences 7.4-fold up-regulated), of the DNA repair-recombination protein (4 sequences 2.8-fold up-regulated), of the dormancy associated protein (2 sequences 4.5-fold up-regulated), and of the DC1 domain-containing protein (2 sequences 4-fold up-regulated) only hybridized with the late root development enriched probes.

The level of expression of the singleton sequences ranged from 1.1- to 6.3-fold up-regulation. Two of these sequences matched a pentatricopeptide (PPR) repeat-containing protein (1.6-fold up-regulated) and a DC1 domain-containing protein (2.1-fold up-regulated) with different sequences respect those that were clustered. Three ATPases were also detected showing a 1.2-, 1.8-, and 2.4-fold over-expression.

The 110 down-regulated clones represented a total of 65 non-redundant sequences, 36 derived from 80 originally clustered sequences, and 29 from singleton sequences. Of the clustered sequences, 11, 9 and 8 sequences, which mainly hybridized with the 3rd WAE enriched probes, represented an unknown protein (3-fold down-regulated), a jacalin lectin family protein (2.4-fold down-regulated), and a glycine-rich RNA-binding protein (2-fold down-regulated), respectively. Similarly, all sequences of the PRLI factor (2 sequences 8.6-fold down-regulated), of the no apical meristem family protein (2 sequences 5.3-fold down-regulated), of the Bet v I allergen family protein (2 sequences 4.2-fold down-regulated), of the C2 domain-containing protein (4 sequences 2.5-fold down-regulated), of the glycine-rich protein (3 sequences 2.1-fold down-regulated), and of the 60S ribosomal protein (3 sequences 1.6-fold down-regulated) only hybridized with the early root development enriched probes (Tab 4.2). The level of down regulation of the non-clustered sequences ranged from 1.2- to 5-fold differences. Sucrose synthase (SUS1), a gene directly involved in sucrose metabolism, showed a 1.4-fold down-regulation during root development. Nine sequences matched ribosomal subunit proteins ranging from 1.4- to 2-fold down-regulation, while five genes involved in osmotic regulation (Cation-Cl cotransporter; cation efflux family; osmotin–like protein; dehydratation-responsive protein; and a dessication-responsive protein) were strongly down regulated during active sucrose accumulation. Overall, during the first two months of root developmental, expression of genes involved in cell cycle and osmotic regulation were gradually reduced, while genes involved in transcriptional regulation, signal transduction, metabolite transport and energy metabolism were activated. Differently, genes involved in carbohydrate and protein metabolisms were constantly active during root development. A high proportion of down-regulated (37.8 %) and up-regulated (26.5 %) had unknown function.

Arabidopsis thaliana cDNA microarray

Of the 12,580 *A. thaliana* cDNA clones printed on the arrays, 6697 (53.2 %) showed hybridization with probes derived from *B. vulgaris* transcripts, of which 1956 (15.5 %), 2081 (16.5 %), 1799 (14.3 %), and 861 (6.9 %) had low, medium, high, and very high hybridization signals, respectively.

Overall, a total of 332 differentially expressed clones were observed of which 120 (1.8 %) and 212 (3.2 %) clones were up- and down -regulated during late root development respectively, while 6365 (95.0 %) did not show any significant variation of expression between samples.

Of the 120 clones up-regulated at the 7th WAE, only 48 (40.0 %) had transcripts with known

or putative functions, while 28 (23.4 %), 27 (22.5 %), and 17 (14.1 %) matched transcripts with proteins with unknown functions, hypothetical proteins, and with nucleotide with no sequence similarity, respectively. Similarly, of the 212 clones down-regulated at the 7th WAE, only 104 (49.1 %) had transcripts with known or putative functions, while 37 (17.4 %), 50 (23.6 %), and 21 (9.9 %) matched transcripts with proteins with unknown functions, hypothetical proteins, and with nucleotide with no sequence similarity, respectively.

Examining the levels of transcript hybridization, of the 120 clones up-regulated during late root development, only 43 (35.8 %) had transcripts with low hybridization signal, while the other 77 matched transcripts with at least medium hybridization signal (Tab. 4.11). Similarly, of the 212 clones down-regulated during late root development, only 53 (25 %) had transcripts with low hybridization signal, while the other 159 matched transcripts with at least a medium hybridization signal. Considering the 6365 clones that did not show any difference of expression between early and late root developmental stages, similar proportions to differentially expressed genes were observed for clones with low (1860 = 29.2 %), medium (1983 = 31.2 %), and high (1687 = 26.5 %) hybridization signal, but an higher than expected frequency was observed for clones with very high (835 = 13.1 %) hybridization signal. However, of the 861 clones with very high hybridization intensity, 68 showed a too high hybridization signal to be used to calculate relative ratios (saturation of the detector), and gene expression levels were considered as unchanged between samples.

Genes involved in cell cycle, osmotic regulation, signal transduction and stress responses were down-regulated during root development, while genes of carbohydrate, protein and lipid metabolisms and of transcriptional regulation were weekly up-regulated during development. However, the level of gene expression variation between early and late root development was very low, with the majority of transcripts being up-regulated (80.8 %) or down-regulated (84.4 %) between 1.1 and 1.4 fold difference.

Arabidopsis thaliana **oligonucleotide microarray**

The analysis performed using default threshold parameters showed that of the 22,746 probe sets present in the ATH1 Array, 1056 (4.6 %), 228 (1.0%), and 21,462 (94.4 %) were respectively identified as present, marginal and absent. Of those identified as present or marginal, 1234 (96.1 %) did not show any change of expression, while only 46 (3.6 %) and 4 (0.3 %) probe sets showed respectively a decrease and increase of expression during active root sucrose accumulation. Three of the 4 up-regulated genes showed more than a 10-fold difference expression between samples (NADH dehydrogenase D3, cytochrome b6-f complex subunit V, and an unknown protein), while a ribosomal protein L2 showed a 4-fold up-regulation. Differently, the level of variation between the 46 down-regulated genes was not higher than 2.5 fold difference.

Using the non-conventional analysis based on the level of probe set hybridization signals, 845 (3.8 %) and 21,901 (96.2 %) transcripts were respectively identified as present and absent (Tab 4.15). Of those identified as present, only 224 (26.5 %) were present in both 3rd ad 7th WAE samples, while 375 (44.4 %) and 246 (29.1 %) were only present during the early and late phases of root development, respectively. In order to estimate differences of level of gene expression, of the 845 probes identified as present, 274 and 184 were discarded from the analysis because only present at low signals hybridization in one sample. Of the remaining 387 probes, at signal hybridization ratio threshold values of > 1.2 for up-regulated and < -1.2 for down-regulated

genes, 105 and 164 transcripts showed increase and decrease of expression during root development respectively. Only 31.4 % and 37.8 % of the up- and down-regulated genes showed match with transcripts with unknown function, respectively. Differently from the results obtained with default analysis, most (96/105 = 91.4 %) of the differentially up-regulated transcripts showed level of expression smaller than 4-fold difference, and only two of the 4 previously detected up-regulated genes (NADH dehydrogenase D3 and cytochrome b6-f complex, subunit V) were identified as present and up-regulated also with the non-conventional analysis.

Genes involved in cell cycle and signal transduction were down-regulated during root development, while genes of osmotic regulation and of transcriptional regulation were weekly up-regulated during development.

Discussion

Most of the concentric cambium rings that form the majority of the root volume at maturity are formed during the first weeks after emergence (Elliot and Weston, 1993). A chronological analysis of the biological changes that occur during the early root developmental phases in sugar beet, particularly considering the changes in the dynamic of sucrose accumulation, could yield information on overall different productivity between different lines in breeding programs, and could be used as selection parameters. Comparison between sugar beet lines with different sucrose accumulation capability (SR96 > USH20 > C869) showed that these difference could be detected in greenhouse-grown plant as early as after the 7th WAE in 2003 and after the 9th WAE in 2002. These differences, also observed in field-grown plants after three to four months after emergence, were mainly associated with differences in dry matter content more than in sucrose content on the dry matter, as expected by the comparison between elite sugar beet lines. During the field experiment the higher variability observed and the limited number of samples collected masked the expected difference between sugar beet lines. Differences in trait values between 2002 and 2003 greenhouse experiments were probably caused by the later time of transplant (3 weeks difference) in the 2003, which increased the average temperatures and photoperiod during the early root developmental stages. Other than showing the possibility to detect difference between breeding lines, the greenhouse experiment also highlighted that during the first two month after emergence all biochemical pathways involved in sucrose metabolism that influence sucrose content at maturity are already present and fully active. Particularly between the 3rd and the 7th WAE a sharp change of root sucrose content occur at both the dry matter and fresh weight basis, reaching levels comparable to those observed at root maturity. These results are also confirmed by earlier observation of Bergen (1967) and Milford (1973 ad 1988) for field-grown plants and Klotz (2002 and 2004) for greenhouse-grown plants.

Several techniques for a comprehensive time course analysis of differentially expressed genes have been developed during the last years (Kuhn, 2001; Green et al., 2001; Donson et al., 2002). Of these techniques, cDNA-AFLP has mainly been used to study gene expression changes during developmental phases of species where little sequence information was available, such as in Beta vulgaris L. (Bachem et al., 1996; Durrant et al., 2000; Jones et al., 2000; Okamuro et al., 2000; Breyne et al., 2003). Reliability of cDNA-AFLP for genome-wise analysis of gene expression was confirmed in this study by the similarity of results obtained in the two years and by the high ratio of similarity between biological replications analyzed during the second year, particularly considering that differences between biological replications are expected. The results of this research indicated a change of expression of at least 25 % of the genes normally

expressed during the first two month of root development. Gene expression profiles of early developing roots during the first 3 WAE showed the highest number of transcripts expressed, associated with a lack of sucrose accumulation capability. Root tissues in this early developing stage could be mainly involved in cell division and cell expansion, and only a little functional differentiation of these tissues is expected. This stage seems to be gradually changed during the following two weeks (4th and 5th WAE), with the activation of some of the transitional-expressed genes and the deactivation of all the 199 early-expressed genes, causing an overall decrease of expressed transcripts. This transitional phase is characterized by a sharp increase of root sucrose accumulation. Consequently to the deactivation of early-expressed genes, starting at the 5th WAE several lately-expressed genes are activated in the sucrose accumulating root system. Particularly between the 5th to the 6th WAE profiles of expressed genes changed drastically, while similar profiles of expressed genes were observed from the 6th to the 9th WAE. This pattern of gene expression would suggest a subdivision of the first two months of root development in distinct developmental phases: a pre-sucrose accumulation phase of root tissues development during the first 3 weeks where no functional differentiation is observed; and an active sucrose accumulation phase after the 6th WAE where root tissues appear to be already functionally differentiated. A transitional phase when root tissues start to be functionally differentiated and sucrose starts to accumulate occurred from the 4th to the 6th WAE. Because no morphological or physiological markers specifically exist to differentiate juvenile to adult vegetative phases, and because the this transition occurs gradually during development, it would be possible to consider the overall gene expression changes as good indicator of this phase switch, and the functional differentiation of the tissues as further confirmation of this juvenile to adult vegetative phases change in sugar beet root development.

The use of microarray technology to precisely identify genes differentially expressed has been widely used in plants and animals (Lemieux *et al.*, 1998; Lipshutz *et al.*, 1999; Brown and Botstein, 1999; Kuhn, 2001). Oligonucleotide microarrays are generally used to analyze gene expression profiles and characterize genes in species for which genomic sequences information is known (Harmer et al., 2000; Schaffer et al., 2001) but are rarely used for cross-species hybridization studies using transcripts from different species (Kayo et al., 2001). Differently, cDNA-based microarrays can be also used for species for which little genomic sequences information is known, using transcripts selected from cDNA libraries (Wullschleger and Difazio 2003; Casu et al., 2003). This study combined the advantage of the suppression subtractive hybridization technique to only select those transcripts differentially expressed from the cDNA library (Pearson et al., 2001), and the ability to confirm and quantify their relative levels of expression with the cDNA microarray in a species with little sequence information available as in *Beta vulgaris*. The relative abundance of clustered transcripts indicated high levels of expression and possible functional differentiations of the root tissue. However, the majority of the clustered genes represented regulatory genes more than structural genes, implying that several metabolic changes could be active during early root development, as opposed of the variation of only few specific biosynthetic pathways, such as those involved in sucrose biosynthesis and accumulation. Clustered genes down-regulated during root development were mainly involved in the regulation of cell cycle, like ribosomal proteins, or in signal transduction, like for the 8-fold down-regulated PRL1 interacting factor. Interestingly, PRL1 is a negative regulator of the *Arabidopsis* SNF1 kinase, a key regulator of glucose signaling (Bhalerao *et al.*, 1999). Pentatricopeptide (PPR) repeat-containing protein, one of the most abundant clustered transcripts up-regulated at the 7th WAE, is supposed to be have RNA-binding sites and be

involved in RNA processing and stabilization (Small and Peeters, 2000). Functional analysis of singletons confirmed the observations made for the clustered genes. Overall, transcripts involved in cell cycling and osmotic regulation were mainly expressed during the early stage of root development, while genes involved in transcriptional regulation, signal transduction, energy metabolism and metabolites transport were maily expressed during the sucrose accumulating phase of root development. Structural genes involved in proteins, lipids, and particularly carbohydrates metabolism were not found to be differentially expressed between phases, with the exception of sucrose synthase (SUS1), an enzyme directly involved in the catabolism of sucrose, that was weekly over-expressed before sucrose started accumulating in the root. However SUS1 high expression during early root development can be explained considering that SUS1, interacting with cellulose synthase in the plasma membrane, plays also an important function in cell wall biosynthesis (Chourey et al., 1998). Lacking of expression of transcripts for carbohydrate metabolism was also observed in maturing stems in sugarcane, while sugar transporters were mainly expressed (Casu et al., 2003).

Several lines of evidence supported the quality and reliability of the results obtained with the *Beta vulgaris* subtracted library microarray. The majority of the transcripts selected by the subtracted probes hybridized to all four biological replications of the two sugar beet lines, and the levels of gene expression between biological replications were similar between each other. Furthermore, similarity of levels of gene expression observed between independently arrayed clustered sequences support the reliability of results. Also, the suppressive subtractive hybridization demonstrated to be an efficient technique able to select differentially expressed genes, particularly for abundant (clustered) transcripts showing more than 3-fold difference of expression between samples.

Considering the *Arabidopsis thaliana* cDNA microarray experiment, the level of hybridization between *B. vulgaris* probes and *A. thaliana* targets was relatively high (53.2 %). In a similar experiment, Horvath et al. (2003) found levels of hybridization to Arabidopsis cDNA microarray of 23 %, 34% and 47 % for wild oat (*Avena fatua*), poplar (*Populus deltoidsies*) and spurge (*Euphorbia esula*), respectively. In spite of the high level of hybridization, the low number of differentially expressed genes (5 %) and the low folds differences of expression (< 2-fold), indicated the inability of the Arabidopsis-based microarray to precisely identify level of differentially expressed genes during root development in sugar beet. Overall, genes involved in cell cycle, osmotic regulation and stress responses were up-regulated during the early root development, as observed in the *B. vulgaris* cDNA microarray experiment. Differently, sucrose synthase was found to be up-regulated during the sucrose accumulation phase, and two distinct sucrose transporters, SUC1 and SUC2, were found up-regulated during early and late root development, respectively. The level of transcript detection strongly decreased during the Arabidopsis oligonucleotide microarray, when only 3.8 % of the probe sets showed hybridization signals. Affymetrix gene chips are constructed to be analyzed only with conventional and standardized analytical software, and results from data analysis using non-conventional parameters must be considered carefully, and only used as general indicator of differentially expressed genes. Cell cycle related genes were also found to be up-regulated during the early phase of root development, while genes related with energy metabolism and transcriptional regulation were up-regulated during the sucrose accumulation phase. Results obtained with microarrays experiment confirmed the decrease of expressed transcripts between the 3rd and the 7th WAE observed with cDNA-AFLP and the relatively higher proportion of expressed genes in SR96 respect USH20 lines.

Conclusion

(i) Phenotypic analysis of the dynamic of root sucrose accumulation revealed a sharp increase of sucrose content from the 3rd to the 7th week after emergence (WAE) in greenhouse-grown plants. Comparison between sugar beet lines with known difference of sucrose storage capability at maturity showed proportional sucrose content differences after 7 to 9 WAE in greenhouse-grown plants. These differences of sucrose on the fresh weight basis were caused by differences in root dry matter content and not by differences in sucrose content on the dry matter basis. Similar differences observed for greenhouse-grown plants were observed after 13 to 16 WAE in field-grown plants, indicating the potential of early greenhouse screening as selective indicator during breeding programs.

(ii) Gene expression profiles revealed an early root developmental phase during the first 3 WAE characterized by the highest number of expressed transcripts, followed by a transitional phase from the 4th to the 5th WAE where the expression of the early-specific transcripts were decreased, and finally followed by a lately root developmental phase after the 6th WAE characterized by late-specific expressed transcripts and root sucrose content values on dry and fresh weight basis similar to those observed at root maturity. Between the first two and the third phases the highest change in gene expression profiles was observed. Considering the dynamic of sucrose accumulation and of gene expression profiles together, it was hypothesized a developmental change from the juvenile vegetative phase, characterized by functionally undifferentiated tissues, to the adult vegetative phase, characterized by active sucrose accumulating tissues and full activity of those metabolic pathways that influence sucrose content at root maturity, between the 4th and the 6th WAE in the sugar beet root system.

(iii) Genes differentially expressed between developmental phases critical for sucrose accumulation were mainly represented by regulatory genes respect structural genes. Early over-expressed transcripts were mainly associated with cell cycle metabolism and osmotic regulation pathways, while genes up-regulated during active root sucrose accumulation were mainly involved in transcriptional regulation and signal transduction pathways, in energy metabolism and in metabolite transport. Arabidopsis-based microarrays were not completely efficient in detecting hybridizations and change of transcripts expression in *Beta vulgaris* L. gene expression analyses. These findings would increase our understanding on which metabolic pathways regulate the early stages of root development and sucrose accumulation in sugar beet.

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Table 1: 04BB01 Agronomic performance

Entry	Seed Number	Name	Purpose	Sucrose (%)	CJP (%)	T/A	RWSA	RWST	Nitrogen	Poassium	Sodium
42	EL-A011964	B5736	check	20.12	96.88	22.4	8753.1	389.1	102.9	1705.6	113.5
15	EL-A013507	OS-EL0204smrlP	1	18.35	95.35	24.4	8551.1	349.3	125.7	1758.9	185.2
10	EL-A013495	MF-SR97smrlP	1	19.11	95.35	23.2	8386.7	363.9	151.2	1764.8	157.6
12	EL-A013501	OB-SR96smrlP	1	18.89	96.15	22.7	8251.7	362.6	101.9	1534.7	159.3
9	EL-A013492	MF-SR96smrlP	1	19.47	95.60	22.0	8165.6	371.6	114.1	1713.1	215.1
36	EL-A012153	SP85657cms X SR94	2	17.80	96.63	22.9	7872.6	343.3	110.6	1549.6	151.8
7	EL-A013475	BI-EL0204smrlP	1	17.85	94.95	23.3	7870.2	338.3	110.3	1773.5	241.5
39	EL-A012159	SP85657cms X SR97	2	17.54	95.43	23.1	7729.4	334.1	118.3	1700.4	150.7
13	EL-A013503	OB-SR97smrlP	1	18.87	96.15	20.5	7436.1	362.2	104.3	1666.3	115.6
20	EL-A013514	RA-01B006smrlP	3	17.53	95.73	21.8	7304.4	335.0	55.7	1679.0	175.4
4	EL-A012172	SR94	check	17.84	95.93	21.2	7239.5	341.7	106.8	1528.8	123.1
14	EL-A013506	OS-95HS2smrlP	1	18.33	95.90	20.6	7221.7	351.0	105.4	1588.3	125.9
28	EL-A013522	RA-SR96smrlP	1	19.76	95.95	19.1	7211.1	378.5	125.7	1666.3	149.5
23	EL-A013517	RA-01B010smrlP	3	18.19	95.05	20.9	7171.4	345.1	127.4	1911.0	198.4
22	EL-A013516	RA-01B009smrlP	3	17.35	95.38	21.5	7091.1	330.3	86.8	1730.6	199.0
16	EL-A013508	OS-SR96smrlP	3	18.31	96.15	20.2	7090.0	351.5	99.1	1550.3	131.1
26	EL-A013520	RA-01B013smrlP	3	19.62	96.05	18.9	7084.0	376.2	76.7	1750.1	155.8
41	EL-A011875	HME17	check	18.33	96.43	19.8	7037.4	352.8	111.3	1398.2	121.9
17	EL-A013510	OS-SR97smrlP	1	18.35	95.93	19.6	6882.6	351.3	94.9	1687.7	112.1
5	EL-A012174	SR97	check	17.22	95.65	20.9	6848.1	328.8	88.6	1617.5	154.7
21	EL-A013515	RA-01B007smrlP	3	18.16	95.88	19.7	6842.9	347.6	60.2	1733.6	150.7
24	EL-A013518	RA-01B011smrlP	3	17.84	95.53	20.0	6791.1	340.1	79.8	1743.3	164.5
29	EL-A013481	BI-USH20smrlP	3	17.87	95.23	19.9	6741.4	339.6	145.6	1689.7	151.8
27	EL-A013521	RA-EL0204smrlP	3	18.23	95.45	19.3	6685.9	347.5	89.3	1780.4	178.3
2	EL-A013523	2xSRrzc,smr (01B024)	4	16.83	94.65	20.2	6414.2	318.1	127.4	1902.2	151.2
37	EL-A012157	FC607cms X SR97	2	17.50	96.43	19.0	6400.5	336.9	47.6	1486.9	127.1
34	EL-A012150	FC607cms X SR94	2	17.93	95.53	18.5	6317.7	341.9	127.8	1659.5	153.0
35	EL-A012152	SP85576cms X SR94	2	16.87	96.28	19.4	6307.6	324.3	49.7	1422.5	129.4
25	EL-A013519	RA-01B012smrlP	3	17.73	95.78	18.5	6281.5	338.9	79.5	1728.7	189.8
18	EL-A013512	RA-01B002smrlP	3	17.27	95.23	19.0	6220.7	328.2	128.8	1657.5	190.3
40	EL-A012858	EL0204	check	17.55	95.30	18.5	6178.6	333.9	135.5	1734.5	132.3
33	EL-A013472	EL50Cerc-smr	5	17.76	96.05	18.1	6169.2	340.5	85.4	1523.9	150.1
32	EL-A012206	ACH185	check	18.12	96.25	17.7	6168.1	348.2	92.4	1463.5	124.2
11	EL-A013499	OB-EL0204smrlP	1	18.01	95.25	17.2	5989.1	342.6	135.5	1748.2	149.5
6	EL-A012186	EL51	check	16.84	95.25	18.2	5862.5	320.1	87.5	1746.2	196.1
1	EL-A013698	HTLTSsmr (00B041)	6	16.20	94.55	18.4	5635.1	305.8	151.6	1764.8	153.5
31	EL-A012198	USH20	check	16.92	95.10	17.4	5570.8	321.2	138.3	1610.7	194.9
30	EL-A013474	BI-SP6822smrlP	3	17.53	95.85	16.3	5469.7	335.5	116.9	1487.9	154.1
19	EL-A013513	RA-01B005smrlP	3	16.44	95.58	16.9	5286.2	313.7	34.0	1683.8	242.1
38	EL-A012158	SP85576cms X SR97	2	17.61	96.43	14.3	4841.4	339.0	55.0	1403.0	143.8
8	EL-A013484	C869 O-type	check	17.31	94.50	10.3	3394.5	326.6	191.8	1929.5	159.3
3	EL-A010074	OW-SR94smrlP	7	16.66	95.20	8.6	2716.8	316.7	122.3	1593.8	191.7
Grand Mean				17.91	95.67	19.4	6677.9	342.1	104.6	1662.0	160.0
LSD (0.05)				1.35	1.02	4.9	1822.6	27.3	73.0	184.7	52.8
CV (%)				6.74	0.88	21.85	24.33	7.13	53.01	10.39	28.52
F value				3.26**	2.29**	2.86**	3.25**	3.31**	1.52*	3.85**	2.85**

Table 2: Germination and emergence data for entries in Test 04BB01.

Entry	Harvested	10-day	20-day	30-day	NaCl	Peroxide
11	33.0	70.8	146.3	185.5	13.8	22.0
13	36.5	54.8	114.5	177.5	6.8	17.3
16	37.0	93.0	127.5	168.5	8.0	12.7
7	38.3	56.5	133.8	166.5	5.8	6.3
9	41.8	63.5	130.5	164.8	9.5	4.7
2	32.8	56.8	132.0	155.8	10.5	20.0
14	34.3	57.5	130.0	150.3	5.5	6.0
1	39.8	42.8	113.0	148.8	13.8	19.7
19	37.5	67.3	118.3	147.0	4.5	14.0
12	40.5	19.3	81.5	144.0	5.3	13.0
26	36.5	49.8	109.3	142.0	3.5	5.7
41	36.8	6.3	112.5	142.0	0.5	10.5
6	35.3	4.8	69.8	136.8	6.0	15.3
27	32.5	55.0	112.8	136.8	2.5	10.3
40	25.3	33.5	116.8	136.0	15.3	15.8
22	35.5	61.3	109.3	135.5	9.8	15.3
17	41.0	72.0	99.0	133.3	4.3	5.7
21	34.5	59.3	112.0	130.3	11.5	15.3
25	39.3	43.0	94.0	130.3	5.8	6.0
23	35.0	44.5	108.8	129.5	3.3	11.7
4	36.8	19.3	92.0	123.8	11.0	19.0
10	37.3	49.5	97.3	123.0	14.0	13.3
15	33.3	45.8	106.0	121.5	9.5	9.3
20	34.3	25.8	76.5	121.5	7.0	15.7
39	36.3	21.8	119.0	120.5	3.3	8.8
18	33.3	44.3	81.0	117.0	3.3	13.3
5	33.5	8.5	73.3	116.0	4.5	18.3
28	36.8	40.0	96.0	115.5	2.0	9.3
42	31.5	45.5	96.0	114.0	4.0	12.3
30	32.5	20.0	77.0	113.0	1.3	20.7
8	24.3	24.3	74.5	108.5	10.3	14.3
29	33.0	13.3	55.0	107.3	1.0	20.7
31	32.8	26.8	80.5	105.8	8.7	16.7
34	31.8	12.0	69.0	103.5	10.3	16.5
37	35.5	14.0	68.8	100.3	7.5	16.3
36	36.8	22.0	78.8	96.5	3.5	11.8
24	31.0	26.0	80.5	94.8	7.5	10.0
35	33.0	11.8	56.3	83.0	3.3	9.5
32	29.0	2.5	42.5	65.0	0.0	13.3
38	26.0	9.3	37.3	58.0	1.8	17.8
33	29.3	17.5	37.8	57.5	1.3	19.0
3	6.0	2.0	7.3	10.0	1.3	2.7
Grand Mean	33.89	36.2	92.7	123.0	6.3	13.2
LSD (0.05)	9.36	25.4	37.1	36.0	3.3	4.1
CV (%)	23.44	75.78	40.16	32.18	71.48	39.79
F value	2.66**	6.21**	4.96**	6.53**	11.55**	12.07**

Entry	Polarimetry	EFA	NIR	Water	Non-sucrose DM	Suc/DM
1	16.20	16.27	16.52	0.764	0.236	0.688
2	16.83	17.12	16.91	0.761	0.239	0.704
3	16.66	18.25	17.92	0.763	0.237	0.702
4	17.84	18.23	17.50	0.754	0.246	0.724
5	17.22	18.37	18.63	0.756	0.244	0.706
6	16.84	16.51	17.10	0.759	0.241	0.699
7	17.85	16.37	16.44	0.763	0.237	0.754
8	17.31	17.43	17.58	0.752	0.248	0.697
9	19.47	19.94	19.04	0.739	0.261	0.745
10	19.11	19.07	18.54	0.744	0.256	0.745
11	18.01	18.90	17.39	0.757	0.243	0.740
12	18.89	18.97	18.36	0.742	0.258	0.732
13	18.87	18.62	18.74	0.743	0.257	0.736
14	18.33	18.98	18.49	0.746	0.254	0.721
15	18.35	18.39	18.34	0.751	0.249	0.737
16	18.31	17.73	18.42	0.743	0.257	0.713
17	18.35	18.93	18.43	0.744	0.256	0.716
18	17.27	16.14	17.08	0.757	0.243	0.709
19	16.44	16.88	16.38	0.763	0.237	0.694
20	17.53	17.46	17.33	0.756	0.244	0.718
21	18.16	17.78	16.98	0.757	0.243	0.749
22	17.35	17.27	16.83	0.757	0.243	0.715
23	18.19	16.56	17.40	0.741	0.259	0.705
24	17.84	16.82	17.62	0.757	0.243	0.733
25	17.73	17.37	16.85	0.763	0.237	0.748
26	19.62	16.81	17.56	0.749	0.251	0.782
27	18.23	18.16	17.57	0.754	0.246	0.725
28	19.76	19.20	18.41	0.747	0.253	0.782
29	17.87	17.39	17.62	0.751	0.249	0.716
30	17.53	16.46	17.66	0.751	0.249	0.705
31	16.92	17.87	17.55	0.759	0.241	0.703
32	18.12	17.20	17.96	0.744	0.256	0.709
33	17.76	18.15	18.67	0.746	0.254	0.699
34	17.93	16.85	17.79	0.751	0.249	0.720
35	16.87	16.78	16.41	0.765	0.235	0.717
36	17.80	16.99	17.56	0.749	0.251	0.709
37	17.50	16.67	16.98	0.754	0.246	0.711
38	17.61	16.06	16.34	0.758	0.242	0.727
39	17.54	17.24	17.17	0.755	0.245	0.715
40	17.55	17.17	17.64	0.760	0.240	0.732
41	18.33	20.10	19.85	0.730	0.270	0.680
42	20.12	20.51	20.11	0.725	0.275	0.731
Grand Mean	17.91	17.71	17.71	0.75	0.25	0.72
LSD (0.05)	1.35	2.60	1.25	0.01	0.01	0.04
CV (%)	6.74	11.08	6.60	1.54	4.66	4.92
F value	3.26**	1.47[ns]	3.91**	4.51**	4.51**	1.90*

Entry	Seed Number	Name	Purpose	Weight (lbs)	Harvested	10-day	20-day	T/A
18	EL-A013491	BI-EL0204smrIP	Intercross (Sucrose)	77.4	29.3	9.3	47.7	28.1
24	EL-A013511	C698CMS x SR	Evaluation	70.9	25.0	1.3	31.7	25.7
13	EL-A013488	MF-00J12smrIP	Intercross (Rhizoc)	67.2	24.7	12.3	34.7	24.4
40	EL-A012172	SR94	Check	66.1	36.7	15.0	95.3	24.0
22	EL-A013481	BI-USH20mrsIP	Intercross (Sucrose)	65.1	28.0	3.7	47.0	23.6
20	EL-A013480	BI-SR97smrIP	Intercross (Sucrose)	63.1	20.0	9.7	29.7	22.9
29	EL-A012346	GH-99J12	Evaluation	60.4	26.3	0.0	40.0	21.9
19	EL-A013478	BI-SR96smrIP	Intercross (Sucrose)	59.3	25.0	6.3	52.7	21.5
30	EL-A011969	WC-J19	Evaluation	59.1	30.0	5.7	51.7	21.5
8	EL-A013700	GH-02B097smr-IP	Intercross (Rhizoc)	59.1	40.7	1.3	76.3	21.4
14	EL-A013489	MF-Trad-ELsmrIP	Intercross (Rhizoc)	57.9	26.7	6.7	43.7	21.0
25	EL-A012176	WC970457	Evaluation	57.8	32.3	33.7	62.3	21.0
7	EL-A013704	GH-02B096smr-IP	Intercross (Rhizoc)	56.8	36.3	2.0	76.3	20.6
42	EL-A012200	EL52	Evaluation	56.8	34.7	20.3	89.7	20.6
11	EL-A013705	GH-02B103smr-IP	Intercross (Rhizoc)	55.6	37.3	3.3	73.3	20.2
4	EL-A007774	GH-01B024smr (SR Rzc)	Evaluation	55.3	30.7	4.7	56.3	20.1
16	EL-A013472	BI-03B031smrIP (96N7)	Intercross (Sucrose)	52.8	23.7	3.7	35.7	19.2
3	EL-A011971	WC-00J12-02	Evaluation	51.1	36.3	8.3	96.0	18.6
2	EL-A012345	GH-00J02smr	Evaluation	50.5	29.7	0.0	27.0	18.3
35	EL-A012344	GH-EL50 (Joe 20 plants cross?)	Evaluation	49.9	24.0	0.0	25.3	18.1
21	EL-A013486	EB-USH20smrIP (Yi-Aph)	Evaluation	49.9	34.7	3.0	30.7	18.1
6	EL-A013703	GH-02B095smr-IP	Intercross (Rhizoc)	49.0	35.0	5.0	61.0	17.8
5	EL-A008061	GH-01B037smr (EL52 cerc)	Evaluation	48.6	20.3	0.3	13.7	17.6
31	EL-A011970	WC-J31	Evaluation	46.9	26.0	1.3	25.3	17.0
15	EL-A013490	MF-FCsmrIP	Intercross (Rhizoc)	46.6	22.0	2.3	29.3	16.9
1	EL-A013699	WC-00B042 (SugXFod)	Evaluation	45.9	36.0	32.7	99.0	16.7
10	EL-A013701	GH-02B100smr-IP	Intercross (Rhizoc)	45.9	28.0	9.0	82.0	16.6
23	EL-A013483	C869 CMS	Evaluation	44.4	24.3	0.0	37.0	16.1
32	EL-A012178	EL38	Evaluation	44.0	25.3	12.7	59.3	16.0
41	EL-A010297	01B001 (SugXFod)	Evaluation	42.6	33.3	18.7	73.3	15.5
12	EL-A013702	GH-02B153smr (Trad-EL)	Evaluation	42.5	37.7	2.7	63.3	15.4
34	EL-A012181	EL48	Evaluation	41.5	30.7	7.0	65.3	15.1
37	EL-A007085	GH-SP83303inc	Evaluation	40.1	31.3	4.7	59.0	14.5
17	EL-A013473	BI-00J12smrIP	Intercross (Sucrose)	36.9	6.3	1.7	8.0	13.4
9	EL-A013706	GH-02B098smr-IP	Intercross (Rhizoc)	35.5	26.7	0.0	26.3	12.9
28	EL-A012859	WC-J24	Evaluation	34.0	14.3	2.7	23.7	12.3
33	EL-A013473	BI-00J12smrIP	Evaluation	28.6	5.7	2.0	6.0	10.4
39	EL-A010088	OW-USH20smrIP	Evaluation	26.9	5.7	0.0	7.3	9.8
27	EL-A010068	OW-EL51smrIP	Evaluation	24.2	7.3	0.0	9.0	8.8
36	EL-A010071	OW-SR-MIXsmrIP	Evaluation	17.7	5.0	0.0	5.0	6.4
38	EL-A010084	OW-SR93smrIP	Evaluation	16.8	8.7	1.0	13.7	6.1
26	EL-A010075	OW-RM10smrIP(rzm)	Evaluation	0.0	0.0	0.0	0.0	0.0
	Grand Mean			47.64	25.28	6.05	44.99	17.71
	LSD (0.05)			11.21	6.84	5.94	19.32	4.12
	CV (%)			34.5	43.53	140.02	64.55	30.34
	F value			15.22**	18.77**	14.28**	16.12**	11.65**

Table 4: 00BB02 Evaluation plots

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Entry	Seed Number	F4 Parent	Beet Weight	Sucrose FW (%)	Water (%)	Dry Matter (%)	Sucrose DM (%)
1	EL-A013707	03B109-01	0.32	16.6	75.6	24.4	67.9
2	EL-A013708	03B109-02	1.59	15.8	76.8	23.2	68.1
3	EL-A013709	03B109-03	0.91	14.9	77.3	22.7	65.4
4	EL-A013710	03B109-04	1.57	15.6	77.1	22.9	67.7
5	EL-A013711	03B110-01	0.32	18.6	74.2	25.8	71.8
6	EL-A013712	03B110-02	1.59	16.6	76.5	23.5	70.6
7	EL-A013713	03B115-03	1.17	14.0	78.3	21.7	64.4
8	EL-A013714	03B115-04	1.69	15.4	77.3	22.7	67.9
9	EL-A013715	03B118-02	0.86	16.0	77.4	22.6	70.9
10	EL-A013716	03B118-03	1.24	15.3	77.9	22.1	69.5
11	EL-A013717	03B121-02	1.73	17.9	75.5	24.5	73.2
12	EL-A013718	03B123-04	1.10	17.8	75.1	24.9	71.6
13	EL-A013719	03B124-01	0.68	17.4	75.5	24.5	70.9
14	EL-A013720	03B124-02	1.69	16.7	75.2	24.8	67.1
15	EL-A013721	03B124-04	1.84	16.4	75.9	24.1	68.3
16	EL-A013722	03B126-01	3.17	17.2	75.2	24.8	69.3
17	EL-A013723	03B126-02	0.49	19.0	75.2	24.8	76.7
18	EL-A013724	03B126-04	2.32	17.6	77.0	23.0	76.5
19	EL-A013725	03B127-01	1.23	17.7	76.5	23.5	75.4
20	EL-A013726	03B127-02	2.34	17.4	76.0	24.0	72.5
21	EL-A013727	03B127-03	0.00	nd	nd	nd	nd
22	EL-A013728	03B127-04	0.96	18.3	76.0	24.0	76.3
23	EL-A013729	03B127-05	1.52	16.4	76.8	23.2	71.0
24	EL-A013730	03B129-01	1.22	17.1	76.5	23.5	72.9
25	EL-A013731	03B129-03	1.50	17.0	77.1	22.9	74.3
26	EL-A013732	03B129-04	1.75	16.3	77.2	22.8	71.2
27	EL-A013733	03B129-02	1.46	16.1	76.7	23.3	69.2
28	EL-A013734	03B135-02	1.42	15.7	77.1	22.9	68.7
29	EL-A013735	03B135-03	2.33	16.5	77.1	22.9	71.8
30	EL-A013736	03B136-03	1.75	18.4	75.8	24.2	76.0
31	EL-A013737	03B138-04	1.34	18.6	75.9	24.1	77.0
32	EL-A013738	03B139-01	2.08	16.1	77.4	22.6	71.5
33	EL-A013739	03B140-01	1.35	17.7	75.8	24.2	73.0
34	EL-A013740	03B140-02	1.51	17.0	76.1	23.9	71.1
35	EL-A013741	03B140-03	1.35	17.4	76.4	23.6	73.6
36	EL-A013742	03B140-04	2.14	16.9	75.9	24.1	70.1
37	EL-A013743	03B141-01	1.29	17.4	75.1	24.9	69.8
38	EL-A013744	03B141-02	0.90	19.0	74.0	26.0	73.1
39	EL-A013745	03B144-03	1.40	15.3	77.5	22.5	67.5
40	EL-A013746	03B147-01	1.65	17.2	75.7	24.3	70.7
41	EL-A013747	03B147-02	1.27	17.4	75.6	24.4	71.4
42	EL-A013748	03B147-03	1.59	17.8	75.5	24.5	72.8
43	EL-A013749	03B147-04	1.27	16.4	76.4	23.6	69.3
44	EL-A013750	03B149-02	1.42	17.3	76.1	23.9	72.6
45	EL-A013751	03B149-03	1.75	16.4	76.9	23.1	70.9
46	EL-A013752	03B149-04	1.08	16.5	76.9	23.1	71.3
47	EL-A013753	03B150-04	1.98	14.8	77.9	22.1	67.0
48	EL-A013754	03B152-01	1.45	16.0	76.1	23.9	66.7
49	EL-A013755	03B153-02	3.58	15.6	77.0	23.0	68.0
50	EL-A013756	03B153-03	1.71	17.0	76.2	23.8	71.5
51	EL-A013757	03B158-02	2.67	16.2	77.4	22.6	71.4
52	EL-A013758	03B158-03	2.42	16.3	76.8	23.2	70.0
53	EL-A013759	03B159-01	1.25	17.0	76.7	23.3	72.7
54	EL-A013760	03B160-04	1.37	14.6	77.9	22.1	66.3
55	EL-A013761	03B161-01	1.25	19.0	74.2	25.8	73.6
56	EL-A013762	03B161-03	1.17	19.8	74.4	25.6	77.1
57	EL-A013763	03B161-04	1.61	16.5	76.3	23.7	69.8
58	EL-A013764	03B163-02	1.81	15.6	78.0	22.0	70.9
59	EL-A013765	03B166-02	0.97	16.7	77.0	23.0	72.5
60	EL-A013766	03B166-04	1.07	18.5	75.6	24.4	75.9

61	EL-A013767	03B168-01	2.35	15.0	77.4	22.6	66.1
62	EL-A013768	03B169-01	1.19	18.4	75.0	25.0	73.6
63	EL-A013769	03B169-02	2.48	18.4	74.7	25.3	72.6
64	EL-A013770	03B169-04	0.79	20.5	72.4	27.6	74.4
65	EL-A013771	03B171-01	2.06	16.8	77.8	22.2	75.4
66	EL-A013772	03B171-02	1.13	17.0	76.0	24.0	70.9
67	EL-A013773	03B171-03	1.43	17.8	76.1	23.9	74.3
68	EL-A013774	03B172-04	2.17	16.0	77.4	22.6	70.5
69	EL-A013775	03B173-01	0.71	17.7	75.1	24.9	71.2
70	EL-A013776	03B174-04	1.82	15.1	78.0	22.0	68.3
71	EL-A013778	03B179-02	0.88	17.1	76.7	23.3	73.2
72	EL-A013779	03B179-03	1.27	16.0	77.5	22.5	71.2
73	EL-A013780	03B180-03	1.38	15.7	78.2	21.8	72.2
74	EL-A013781	03B181-01	1.24	16.5	77.1	22.9	72.3
75	EL-A013782	03B181-02	1.93	17.0	76.9	23.1	73.3
76	EL-A013783	03B181-04	1.03	17.8	75.9	24.1	73.7
77	EL-A013784	03B183-04	1.55	18.3	75.6	24.4	74.9
78	EL-A013785	03B184-01	1.80	17.1	77.0	23.0	74.6
79	EL-A013786	03B184-02	1.69	17.1	76.8	23.2	73.8
80	EL-A013787	03B186-01	1.08	17.7	75.5	24.5	72.4
81	EL-A013788	03B187-01	1.85	16.2	76.7	23.3	69.6
82	EL-A013789	03B189-01	0.85	18.1	74.7	25.3	71.5
83	EL-A013790	03B189-02	1.24	17.9	75.0	25.0	71.5
84	EL-A013791	03B189-03	1.62	17.7	75.2	24.8	71.3
85	EL-A013792	03B189-04	1.13	17.6	75.5	24.5	71.9
86	EL-A013793	03B191-02	1.18	17.7	74.6	25.4	69.8
87	EL-A013794	03B195-01	1.12	18.1	75.9	24.1	75.3
88	EL-A013795	03B195-02	1.35	18.6	74.9	25.1	73.8
89	EL-A013796	03B195-03	1.53	18.7	74.5	25.5	73.4
90	EL-A013797	03B195-04	1.11	17.8	74.9	25.1	71.1
91	EL-A013798	03B197-01	2.14	15.6	77.5	22.5	69.3
92	EL-A013799	03B198-01	1.39	16.7	76.7	23.3	71.7
93	EL-A013800	03B198-02	2.95	14.8	77.4	22.6	65.6
94	EL-A013801	03B201-01	3.42	16.1	77.3	22.7	70.6
95	EL-A013802	03B201-02	2.41	15.6	77.9	22.1	70.6
96	EL-A013803	03B205-01	2.30	16.4	76.5	23.5	69.7
97	EL-A013804	03B205-02	2.15	15.6	77.3	22.7	68.4
98	EL-A013805	03B207-01	2.15	15.9	77.3	22.7	70.0
99	EL-A013806	03B207-02	2.54	15.4	77.3	22.7	67.6
100	EL-A013807	03B211-02	2.31	16.4	76.3	23.7	68.9
101	EL-A013808	03B212-01	1.83	16.8	75.8	24.2	69.4
102	EL-A013809	03B212-02	1.93	18.3	74.8	25.2	72.7
103	EL-A013810	03B212-03	2.29	16.1	77.0	23.0	69.8
104	EL-A013812	03B214-01	1.48	18.0	75.9	24.1	74.7
105	EL-A013813	03B214-02	1.25	16.8	76.1	23.9	70.2
106	EL-A013814	03B214-03	1.76	18.5	74.1	25.9	71.4
107	EL-A013815	03B216-02	1.30	16.8	75.9	24.1	69.6
108	EL-A013816	03B216-03	0.80	16.9	75.7	24.3	69.5
109	EL-A013817	03B216-04	1.08	17.2	75.2	24.8	69.3
110	EL-A013818	03B217-01	2.04	14.7	77.3	22.7	64.7
111	EL-A013819	03B217-02	1.71	14.7	77.9	22.1	66.6
112	EL-A013820	03B217-04	1.97	12.5	79.1	20.9	59.8
113	EL-A013821	03B220-01	1.40	18.8	75.3	24.7	75.8
114	EL-A013822	03B222-01	1.87	16.9	76.5	23.5	71.9
115	EL-A013823	03B221-02	1.61	17.4	76.5	23.5	74.0
116	EL-A013824	03B221-03	2.25	14.9	77.4	22.6	65.8
117	EL-A013825	03B221-04	1.27	14.8	78.8	21.2	70.0
118	EL-A013826	03B221-01	1.99	14.9	78.7	21.3	70.0
119	EL-A013827	03B222-03	1.27	16.7	77.6	22.4	74.7
120	EL-A013828	03B222-04	1.01	16.9	76.1	23.9	70.8
121	EL-A013829	03B225-01	0.82	18.7	75.0	25.0	75.3

			Beet Weight	Sucrose FW (%)	Water (%)	Dry Matter (%)	Sucrose DM (%)
122	EL-A013830	03B225-02	0.99	17.4	76.8	23.2	74.9
123	EL-A013831	03B225-03	1.47	17.5	77.0	23.0	75.9
124	EL-A013832	03B225-04	1.39	15.3	78.4	21.6	71.0
125	EL-A013833	03B229-01	1.87	15.3	77.7	22.3	68.4
126	EL-A013834	03B230-02	1.62	17.9	75.3	24.7	72.6
127	EL-A013835	03B240-04	1.89	15.7	77.8	22.2	70.3
128	EL-A013836	03B242-02	2.22	16.8	77.3	22.7	73.9
129	EL-A013837	03B243-04	2.21	16.6	77.0	23.0	72.2
130	EL-A014108	03B139-02	1.66	18.0	75.5	24.5	73.5
131	EL-A014109	03B144-02	1.28	15.8	76.7	23.3	67.4
132	EL-A014110	03B161-02	2.02	16.6	76.8	23.2	71.3
133	EL-A014111	03B176-04	1.96	16.8	76.6	23.4	71.5
134	EL-A014112	03B194-01	0.53	19.5	74.3	25.7	75.7
135	EL-A014113	03B199-01	2.38	16.1	76.9	23.1	69.7
136	EL-A014114	03B216-01	1.12	17.1	76.0	24.0	71.4
137	EL-A014115	03B217-03	1.50	14.8	78.3	21.7	67.9
138	EL-A014116	03B219-01	1.30	15.3	77.1	22.9	66.8
139	EL-A014117	03B227-04	1.32	15.8	78.1	21.9	72.3
140	EL-A014118	03B229-02	1.67	14.8	77.6	22.4	65.9
141	EL-A014119	03B230-01	1.80	16.7	76.0	24.0	69.5
142	EL-A014120	03B230-03	0.97	15.6	76.5	23.5	66.6
143	EL-A014121	03B235-01	2.28	13.2	79.0	21.0	62.8
144	EL-A014122	03B235-03	0.00	nd	nd	nd	nd
145	EL-A014123	03B235-04	2.32	16.4	76.4	23.6	69.4
146	EL-A014124	03B236-01	1.48	14.9	78.0	22.0	67.7
147	EL-A014125	03B237-01	1.54	14.9	77.8	22.2	67.1
148	EL-A014126	03B237-02	1.28	16.2	76.8	23.2	69.5
149	EL-A014127	03B238-01	1.64	15.6	77.1	22.9	68.2
150	EL-A014128	03B240-01	1.82	15.2	77.4	22.6	67.3
151	EL-A014129	03B240-02	1.54	14.5	78.9	21.1	68.4
152	EL-A014130	03B240-03	1.14	15.4	76.6	23.4	65.8
153	EL-A014131	03B242-01	1.69	17.6	75.8	24.2	72.9
154	EL-A014133	03B243-02	1.28	16.7	76.3	23.7	70.4
155	EL-A014107	03B118-01/0:	1.41	17.5	76.1	23.9	73.3
156	EL-A014140	03B192-04	1.03	18.6	74.3	25.7	72.3
157	EL-A014141	03B206-04	1.14	18.3	74.9	25.1	72.8
	commercial	C913	3.10	17.2	75.9	24.1	71.1
	commercial	C913	1.46	20.4	73.3	26.7	76.1
	commercial	C913	3.19	19.3	74.5	25.5	75.9

			Beet Weight	Sucrose FW (%)	Water (%)	Dry Matter (%)	Sucrose DM (%)
Grand Mean			1.58	16.77	76.42	23.58	70.99
LSD (0.05)			0.63	1.66	1.17	1.17	4.92
CV (%)			45.50	10.72	1.91	6.19	6.57
F value			6.16**	5.08**	8.04**	8.04**	2.93**

Table 6: Stand counts of entries tested in Test 04BB03 (Disease nursery).

Entry	Seed Number	Female parent	# of Plots	Stand counts			
				11-day	18-day	25-day	109-day
1	Hero	03B081 (HERO)	12	13.6	39.8	30.8	18.7
3	EL-A013485	03B033	8	2.0	7.8	2.6	2.4
4	EL-A013698	00B041	4	45.5	56.8	40.8	22.5
5	EL-A013699	00B042	4	33.5	26.0	14.0	10.3
6	EL-A011969	99J19-00 (mm SR inc)	4	32.5	45.8	39.3	27.8
7	EL-A011970	99J31-00 (mm SR inc)	4	14.0	15.0	12.8	10.5
8	EL-A011971	00J12-02	4	27.0	35.5	22.0	20.0
9	EL-A013517	03B026	4	33.0	45.5	32.3	19.0
10	EL-A013520	03B029	4	39.5	50.3	29.0	21.8
11	EL-A013521	03B047	4	22.5	31.5	16.5	12.0
12	EL-A007774	01B024 MIX	4	22.3	37.8	31.3	21.3
13	EL-A013512	03B021	4	43.0	52.3	34.0	25.3
14	EL-A013513	03B022	4	18.3	23.0	13.5	13.8
15	EL-A013514	03B023	4	34.8	40.8	25.3	18.0
16	EL-A013515	03B024	4	36.0	44.5	27.0	19.8
17	EL-A013516	03B025	4	29.8	43.8	21.8	19.0
18	EL-A013518	03B027	4	25.8	31.3	20.8	16.5
19	EL-A013519	03B028	4	42.0	51.8	35.3	21.0
20	EL-A013522	03B053	4	29.0	39.8	33.5	17.0
21	EL-A005368	SP6822-10	2	11.0	7.5	11.5	8.0
22	EL-A005392	Z430-1 ms	4	13.8	22.5	13.5	9.3
23	EL-A005394	Z430-B ms	2	37.5	39.5	28.0	19.5
24	EL-A005614	98B017-03	2	39.0	37.0	19.5	20.0
25	EL-A010075	96RM10-2 (ow steck 98 row 17)	2	0.0	4.0	2.0	1.5
26	EL-A010068	96RR (ow steck 98 row 1)	2	0.5	5.5	4.5	3.5
27	EL-A010071	MIX (ow steck 98 row 2)	2	1.0	2.0	1.5	2.5
28	EL-A010084	SR93 (ow steck 98 row 13)	2	1.5	8.5	5.0	8.5
29	EL-A010074	SR94 (ow steck 98 row 7)	4	0.3	1.0	0.3	1.3
30	EL-A010088	USH20 (ow steck 98)	2	1.5	0.5	0.5	0.5
31	EL-A012172	WC960448 = SR94	2	54.5	64.0	32.5	22.0
32	EL-A012174	WC960452 = 94HS25 = SR97	2	25.5	38.0	33.0	17.5
33	EL-A012176	96RHS21-7	2	42.5	49.0	37.0	20.5
34	EL-A012178	EL38	2	28.0	17.0	10.5	9.0
35	EL-A012181	EL48 (82B10-00)	2	26.0	37.5	27.0	19.0

Table 6: 04BB03 Aphanomyces 1

36	EL-A012186	96RR = EL51	3	30.7	64.7	52.0	27.0
37	EL-A012859	98J24-01	2	35.0	34.5	31.0	18.5
38	EL-A013472	03B031	2	18.5	48.0	38.0	20.5
39	EL-A013473	03B036	2	1.5	2.5	1.5	2.5
40	EL-A013474	03B041	2	26.0	42.0	27.0	19.0
41	EL-A013475	03B046	4	19.0	33.5	23.5	15.8
42	EL-A013478	03B051	2	21.0	47.0	19.0	23.0
43	EL-A013480	03B057	2	12.5	20.0	17.0	9.5
44	EL-A013481	03B062	2	28.0	38.0	25.0	18.5
45	EL-A013486	03B061	2	19.5	22.0	14.0	12.5
46	EL-A013488	03B016	2	14.0	25.0	22.0	19.0
47	EL-A013489	03B018	2	25.5	34.0	24.5	21.5
48	EL-A013490	03B019	2	12.5	21.0	16.5	10.5
49	EL-A013491	03B049	4	22.5	37.8	29.8	22.8
50	EL-A013492	03B051	2	23.0	31.5	23.5	11.0
51	EL-A013495	03B056	2	35.5	63.5	42.5	23.5
52	EL-A013499	03B046	2	37.5	64.5	50.5	24.0
53	EL-A013501	03B051	2	28.5	47.5	28.0	20.0
54	EL-A013503	03B056	2	40.5	67.0	55.0	31.0
55	EL-A013506	03B030	2	15.0	26.0	18.0	14.0
56	EL-A013507	03B050	2	17.0	34.0	23.0	20.0
57	EL-A013508	03B051	2	34.0	61.5	45.5	32.0
58	EL-A013510	03B057	2	28.0	42.5	27.5	20.5
59	EL-A013523	99EL0204 = EL0204	4	28.5	64.0	49.3	32.0
60	EL-A012858	03B017	4	28.8	38.8	33.0	22.0
62	EL-A007739	00B016-01	2	2.5	6.5	3.0	3.5
63	EL-A007747	00B019-01	2	8.0	26.0	21.0	18.0
64	EL-A007748	00B019-03	2	7.0	17.5	15.0	5.5
65	EL-A013487	03B064	2	11.0	27.0	19.0	16.0
66	EL-A013707	02B092	2	3.5	42.0	19.5	22.0
67	EL-A005249	6869-01ms	1	9.0	28.0	21.0	13.0
68	EL-A005271	6869-15ms	1	11.0	51.0	40.0	28.0
69	EL-A005283	6869-27ms	1	0.0	14.0	11.0	10.0
70	EL-A005825	6869-24ms	1	12.0	26.0	31.0	22.0
71	EL-A005824	98B035-41ms	1	29.0	33.0	28.0	10.0
72	EL-A005834	98B035-57ms	1	20.0	31.0	25.0	17.0
73	EL-A005899	98B043-04ms	1	4.0	17.0	30.0	10.0

Table 6: 04BB03 Aphanomyces 2

			Rep				
74	EL-A006695	00B016-01	1	4.0	42.0	21.0	18.0
75	EL-A006747	00B016-02	1	9.0	17.0	42.0	9.0
76	EL-A006748	00B016-03	1	9.0	26.0	25.0	18.0
77	EL-A006751	00B019-05	1	3.0	10.0	10.0	6.0
78	EL-A006753	00B020-03	1	21.0	44.0	45.0	30.0
79	EL-A007045	99EL 8,9,10-1 x 98B040-48 (SF)	1	0.0	21.0	17.0	15.0
80	EL-A007076	99EL07-2	1	0.0	3.0	0.0	2.0
81	EL-A013484	03B065	4	5.5	19.0	15.0	7.0
82	EL-A013705	02B103	4	24.3	56.3	43.0	23.3
83	EL-A012870	3VS2084	2	19.5	41.0	30.5	17.5
84	EL-A012871	3AC555	2	28.0	57.5	44.5	29.5
85	EL-A012869	8BA4522	2	0.5	4.0	5.5	4.0
86	EL-A012872	016AB	2	11.0	51.0	35.0	29.5
2A	Hero sibs	03B080	1	8.0	39.0	24.0	14.0
2B	Hero sibs	03B083	1	12.0	23.0	17.0	10.0
2C	Hero sibs	03B088	1	10.0	21.0	12.0	11.0
2D	Hero sibs	03B089	1	29.0	45.0	28.0	22.0
2E	Hero sibs	03B090	1	13.0	43.0	25.0	15.0
2F	Hero sibs	03B092	1	12.0	24.0	9.0	12.0
2G	Hero sibs	03B095	1	12.0	31.0	11.0	16.0
2H	Hero sibs	03B102	1	10.0	28.0	12.0	17.0
2I	Hero sibs	03B103	1	11.0	61.0	37.0	26.0
2J	Hero sibs	03B106	1	45.0	81.0	50.0	30.0
2K	Hero sibs	2004 MIX-1 (03B096&03B101)	1	8.0	34.0	36.0	28.0
2L	Hero sibs	03B097	1	4.0	7.0	15.0	12.0
border		03B017 (2X SR Rzt)	1	22.0	50.0	50.0	26.0
	Grand Mean			21.14	34.47	24.58	16.68
	LSD (0.05)			15.51	19.45	18.62	10.13
	CV (%)			74.1	58.9	64.4	57.4
	F value			3.41**	3.75**	3.15**	2.81**

Table 6: 04BB03 Aphanomyces 3

SUGARBEET RESEARCH

2004 REPORT

Section E

Molecular Plant Pathology Laboratory
Agricultural Research Service
United States Department of Agriculture
Beltsville, Maryland

Dr. Ann C. Smigocki, Research Geneticist
Dr. David Kuykendall, Plant Pathologist

CONTENTS

Improvement of root maggot and disease resistance in sugarbeet

BSDF Project 811

Ann C. Smigocki

Introduction:

Improvements in various sugar beet traits such as sugar yield have been achieved by traditional breeding programs, however, many significant disease and pest problems have not been solved. The sugar beet root maggot is the most devastating insect pest in U.S. sugar beet production. Insecticides are the most efficacious means of control, but alternate and environmentally friendly control measures are needed.

Progress: (selected abstracts)

PUTHOFF, DAVID P. and ANN C. SMIGOCKI, USDA-ARS Molecular Plant Pathology Laboratory, BARC-West B004 10300 Baltimore Ave., Beltsville, MD 20705. **Sugar beet genes regulated by sugar beet root maggot (*Tetanops myopaeformis*) infestation.**

We are employing the Suppressive Subtractive Hybridization (SSH) method to identify genes regulated in sugar beet roots after sugar beet root maggot (SBRM) larval feeding. Two beet genotypes are being used in this study: F1010, a susceptible line, and F1016, a moderately resistant line. Root and hypocotyl tissues infested with SBRM for 24 and 48 h were compared to uninfested tissues within each genotype. SSH was conducted between the two genotypes in order to identify genes reciprocally regulated (up-regulated in 1 genotype while down-regulated in the other). Identifying genes from both lines will not only yield a class of genes potentially involved in the defense response of sugar beet to SBRM, but also allows the elucidation of a class of genes associated with the susceptible response. These two classes of genes will be useful in developing future control methods. To date, over

1000 cDNA fragments have been isolated for further characterization that includes, confirmation of differential expression, sequencing, full length cDNA cloning and expression profiling following various plant stresses. Candidate genes identified from all or any of the subtractions will lead to a better understanding of the mechanisms of infestation, resistance and susceptibility.

IVIC-HAYMES, SNEZANA D.[1], MARK BOETEL[2], LARRY G. CAMPBELL[3], ROBERT DREGSETH[2] and ANN C. SMIGOCKI[1]. [1]USDA, ARS, Molecular Plant Pathology Laboratory, 10300 Baltimore Ave, Beltsville, MD 20705, [2]North Dakota State University, Department of Entomology, Hultz Hall, Fargo, ND 58105, and [3]USDA, ARS, Northern Crop Science Laboratory, Fargo, ND 58105.
An *in vitro* sugar beet root maggot (*Tetanops myopaeformis*) feeding assay.

An *in vitro* system was established to study interactions between sugar beet roots and the sugar beet root maggot (SBRM, *Tetanops myopaeformis* Röder). Sources of root material included hairy root cultures, 14-day-old seedlings and taproots from 1-year-old greenhouse plants. Hairy root cultures were stained in 0.01% safranin or crystal violet and placed on petri plates with ½ strength B5 medium or water-moistened Whatman 3 filter paper or nylon membrane. Seedlings and taproots were placed on nylon membranes. To reduce contamination, benomyl (10 mg/l), cefotaxime (300 mg/l) and carbenicillin (400 mg/l) were added to the plates. First, second and third instar SBRM, obtained either from eggs of laboratory-reared flies or from soil samples collected from infested sugar beet fields, were placed on the roots. Evidence of SBRM feeding included severed roots and safranin or crystal violet in the frass or intestinal tracts of insects. Some larvae survived for more than 50 days on the roots. This bioassay will be useful for rapid screening of newly developed SBRM resistant sugar beet germplasm, chemical control agents or biocontrol organisms.

Insecticidal plant extracts and spores of a biocontrol fungus, *Syngliocladium tetanopsis*, are currently being evaluated by this assay.

SNEZANA D. IVIC-HAYMES AND ANN C. SMIGOCKI. Molecular Plant Pathology Laboratory, United States Department Of Agriculture, Agricultural Research Service, Beltsville, MD 20705, USA **Biolistic transformation of highly regenerative sugar beet (*Beta vulgaris* L.) leaves.**

Leaves of greenhouse-grown sugar beet (*Beta vulgaris* L.) plants that were first screened for high regeneration potential were transformed via particle bombardment with the *uidA* gene fused to the osmotin or proteinase inhibitor II gene promoter. Stably transformed calli were recovered as early as 7 weeks after bombardment and GUS positive shoots regenerated 3 months after bombardment. The efficiency of transformation ranged from 0.9-3.7% and stable integration of the *uidA* gene into the genome was confirmed by Southern blot analysis. The main advantages of direct bombardment of leaves to regenerate transformed sugar beet include 1) a readily available source of highly regenerative target tissue, 2) minimal tissue culture manipulation before and after bombardment, and 3) the overall rapid regeneration of transgenic shoots.

ANN C. SMIGOCKI DENNIS WILSON. Molecular Plant Pathology Laboratory, Agricultural Research Service, U.S. Department of Agriculture, Beltsville, MD 20705, USA. **Pest and disease resistance enhanced by heterologous suppression of a *Nicotiana plumbaginifolia* cytochrome P450 gene *CYP72A2***

The functional role of the *Nicotiana plumbaginifolia* cytochrome P450 gene *CYP72A2* was investigated in transgenic plants. *N. tabacum* plants transformed with a sense or antisense *CYP72A2* construct exhibited reduced heights, branched stems, smaller leaves

and deformed flowers. Western blot analysis revealed reduced levels of a 58kDa protein corresponding to CYP72A2, suggesting that the *CYP72A2* homolog was suppressed in the sense and antisense plants. Transgenic plants had increased resistance to *Manduca sexta* larvae that consumed about 35 to 90% less of transgenic vs. control leaves. A virulent strain of *Pseudomonas syringae* pv. *tabaci* induced a disease-limiting response followed by a delayed and decreased development of disease symptoms in the transgenics. *CYP72A2* gene mediated resistance suggests that the plant-pest or -pathogen interactions may have been modified by changes in bioactive metabolite pools.

Summary:

We are developing biotechnological approaches for control of the root maggot. In one of the approaches, we identified the digestive enzymes in the maggot's midgets and demonstrated that their activity was blocked with specific inhibitors purified from different plants (1). We isolated the genes for these inhibitors from squash and *Nicotiana* and reengineered them so that they would be produced in sugar beet taproots as a defense mechanism against the root maggot. We also showed a number of ways to induce the production of insecticidal compounds in genetically engineered model plants and demonstrated that these compounds were lethal to the root maggot larvae (2, 3), suggesting that the production of these toxic compounds in sugar beet may be an effective control strategy for the root maggot To study the interactions between root maggots and sugar beet plants, we established the first laboratory assay that uses seedlings or their corresponding aseptically-maintained root cultures to document the feeding behavior of the root maggot on susceptible and moderately resistant sugar beet varieties (Smigocki et al., 2005, in review). We demonstrated that the root maggot larvae moved away from the resistant roots, whereas they aggregated and fed on the roots of the susceptible variety. The assay generated damaged

taproots that we used to identify genes specifically regulated by root maggot feeding on the susceptible and moderately resistant varieties. About 200 genes were specifically regulated in response to the damage inflicted by the root maggot. Further characterization is ongoing and includes functional analysis of how the genes may be involved in protecting the moderately resistant sugar beet from attack by the insect. Genes deemed as having a potential to impart highest levels of resistance, will be reengineered and introduced into sugar beet in order to develop new sources of resistant germplasm for breeding programs (4, 5).

1. Smigocki, A., S. Ivic, D. Wilson, C. Wozniak, L. Campbell, R. Dregseth, and M. Boetel. Molecular approaches for control of the sugarbeet root maggot. Internat. Inst. Beet Res. Am. Soc. Sugar Beet Tech. Proc., 419-428, 2003.

2. Smigocki, A.C., L.G. Campbell, and C.A. Wozniak. Leaf extracts from cytokinin-overproducing transgenic plants are lethal to sugar beet root maggot (*Tetanops myopaeformis*) larvae. J. Sugar Beet Res. 40 (4): 197-207, 2003.

3. Smigocki, A. and D. Wilson. Pest and pathogen resistance enhanced by heterologous suppression of a cytochrome P450 gene *CYP72A2*. Biotech. Lett. 26: 1809-1814, 2004.

4. Ivic-Haymes, S. and A. Smigocki. Biolistic transformation of highly regenerative sugar beet (*Beta vulgaris* L.) leaves. Plant Cell Rep. 23:699-704, 2005.

5. Ivic-Haymes, S. and A. Smigocki. Regeneration potential of sugar beet leaf callus. In Vitro Cell. & Develop. Biol. 41 (4): in press, 2005.

CFP Transgenic Sugar Beet Seed Produced for Genetic Crosses
L. David Kuykendall, Lee Panella, and R.T. Lewellen
Beltsville, Maryland, Ft. Collins, Colorado, and Salinas, CA

BSDF Annual Report for 2004

Cercospora-induced leafspot disease is a serious problem for sugar beet production in most of the U.S. growing regions. Since conventional plant breeding has thus far successfully produced only moderate leafspot resistance, a biotechnological approach has been pursued since 1998. The Beltsville plant pathology laboratory introduced the cercosporin toxin export gene, *cfp*, from *Cercospora kikuchii* into *Beta vulgaris* L. using inter-kingdom bacterial conjugation; this research accomplishment was documented in a scientific report originally published in Biotechnology Letters, in 2003, in which the transformed plants were described as having been derived from the biotechnology clone 'Rel-1', developed by the late J. Saunders, ARS/USDA in East Lansing, Michigan. Stability of the CFP gene insertion into the genome was verified by Polymerase Chain Reaction (PCR), and, of course, contamination with bacteria excluded. In 2004, a second scientific research report was published in Biotechnology Letters; in this report our Beltsville lab, teamed with the ARS *Cercospora* research lab in Raleigh, NC, described the expression of both transcribed RNA and translated protein in a sugar beet transgenic (PT7#12) carrying CFP by a combination of reverse transcriptase PCR and Western blot antigen detection methodology, both earlier described in a 2003 BSDF annual report. In 2004, a number of viable seeds were produced, even though leafspot evaluation was not practical since very early bolting prevented the development of vegetative plants of sufficient size and maturity, the project moved into a collaborative interaction with the Sugar Beet Research Unit in Fort Collins for the evaluation of the *Cercospora* leafspot resistance of CFP-transgenic sugar beets crossed with different agronomic genotypes with pyramided resistances, namely lines C842 and 9933 from the program of R.T. Lewellen, USDA-ARS, Salinas, CA.

Justification for Research:
The phytopathogenic fungal species *Cercospora beticola* Sacc. has long been known to cause leafspot, the most serious widespread disease of sugar beet in most of the U.S. sugar beet production areas and globally as well. ARS Plant Pathologist John Weiland in North Dakota has recently documented significant genomic diversity among the *Cercospora* fungi that cause leafspot disease in sugar beet (Weiland, 2004). Leafspot destroys primarily the mature, highly photosynthetic leaves, which are necessarily replaced by the growth of new leaves, requiring the utilization of carbohydrate stored in the root, thereby reducing root yield, percent sucrose, and purity of extracted juice. *Cercospora* leaf spot currently is controlled by using moderately disease resistant germplasm, combined with spraying with commercial fungicides. The use of biotechnology to develop greater *Cercospora* leafspot resistant in sugar beet promises a sustainable solution to this problem. That improvement in genetic resistance to this serious pathogen is needed is evidenced by the emergence of mutant *Cercospora* strains that are tolerant to the otherwise highly effective fungicides.

Summary of Literature Review:
Cercospora leafspot has long been serious disease problem in the sugar beet growing areas of the United States where the summers are often hot and humid (Red River Valley, Michigan,

Ohio, and, less often, Great Plains growing areas and California). It has been estimated that a severe epidemic can cause up to a 40% loss of sugar yield (Smith and Martin, 1978; Smith and Ruppel, 1973), or up to a 40% loss in farm revenue (Shane and Teng, 1992).

Resistance to *Cercospora* leafspot has long been a goal of the USDA-ARS sugar beet research program and researchers at Fort Collins, CO long ago developed the techniques necessary to manage screening nurseries in careful way to promote the development of the disease (Ruppel and Gaskill, 1971). Crop rotation with barley, the area's dry climate and low relative humidity allowed this to be done so that the results were rarely tainted with sufficient numbers of other disease-causing organisms. Tolerance to *Cercospora* leafspot is accurately defined as the plant genotype performing well despite symptoms of the disease being present (Fehr, 1987).

Generally the *Cercospora*-resistant germplasm in use today was derived from outcrosses with *B. vulgaris* spp *maritima* to import resistance genes; this seminal plant breeding had been performed in Italy by Munerati (Lewellen, 1992). With this particular genetic source, there are an estimated 4 or 5 genes responsible for *Cercospora* leafspot resistance (Smith and Gaskill, 1970) with broad-sense heritability estimates ranging from 12 to 71% (Bilgen et al., 1969). Narrow-sense heritability estimates of about 24% compared well with realized heritability values, and about 40-60% of the variation environmental (Smith and Ruppel, 1974). Large environmental variation made it difficult to develop resistance through mass selection. Incorporation of *Cercospora* leafspot resistance into varieties with superior agronomic performance was also difficult (Smith and Campbell, 1996) and, therefore, commercial resistant varieties require some fungicide application to provide adequate levels of protection against *Cercospora* (Miller et al., 1994).

A major problem in the development of *Cercospora* leafspot resistant sugar beet is the loss of vigor due to the continual inbreeding (Coons, 1955 and McFarlane, 1971). The use of hybrid varieties has ameliorated this problem to some extent, but seed production on the highly inbred O-type males and CMS females continues to be a problem. This creates an urgent need to continue to the development of a broader genetic base of *Cercospora* leafspot-resistant germplasm. As commercial hybrid parents become more inbred, there must be sufficient diversity in the germplasm base for maximum gain through heterosis. In addition to broadening the genetic base of the commercial sugar beet germplasm, novel genes for resistance to *Cercospora* leafspot resistance might lead to transgression of tolerance to *Cercospora* leafspot, or in other words, the derivation of a population that contains individuals with a phenotype that is beyond the phenotype found in the parents of the population (de Vicente & Tanksley, 1993).

The non-host specific phytotoxic polyketide cercosporin is a lipid-soluble perylenequinone that, upon photoactivation, catalyzes the production of highly reactive oxygen species, principally singlet oxygen (Daub, 1982). Singlet oxygen-catalyzed peroxidation of membrane lipids results in loss of membrane integrity, cytoplasmic leakage, and cell death (Daub & Ehrenshaft, 2000). *Cercospora* hyphae enter the host plant passively through open stomata and grow intercellularly. Toxin-mediated disruption of the cellular membranes of host cells probably provides the pathogen with nutrients for *in situ* growth and sporulation.

Cercosporin-deficient mutants of *C. kikuchii* did not produce lesions on soybean, suggesting cercosporin is an essential virulence factor (Upchurch *et al.* 1991).

Recent studies have focused on identifying genes for resistance to cercosporin in *Cercospora* fungi themselves (Daub & Ehrenshaft 2000). One such resistance mechanism apparently involves the export action of the Major Facilitator (MF)-like protein gene, *CFP*,

which was isolated from *C. kikuchii* (Callahan et al., 1999). Targeted disruption of the *CFP* gene resulted in mutants that lacked virulence on soybean and were inhibited by cercosporin. Cercosporin export was substantially elevated in *CFP* multi-copy strains of *C. kikuchii* that expressed elevated levels of CFP protein (Upchurch *et al*. 2001). Moreover, transgenic expression of *CFP* in the cercosporin sensitive fungus *Cochliobolus heterostrophus* resulted in significantly increased cellular resistance to the toxin (Upchurch *et al*. 2002).

Kanamycin-resistance clones were regenerated *in vitro* following conjugal mating of wounded REL-1 leaf pieces with *Rhizobium radiobacter* carrying pBCFP. Transgenic plants were confirmed by PCR of leaf DNA using *CFP*-specific primers (Kuykendall, et al, 2003). Moreover, vegetatively propagated kanamycin-resistant plants and seed-grown transgenic REL-1 plants stably maintained the ability to produce a DNA product of the approximate size predicted for PCR using the *CFP*-specifc primers.

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Objectives:

1. The evaluation of *Cercospora* leafspot resistance in transgenic sugar beet genotypes, relative to parental germplasm tolerance--- is CFP useful in enhancing leafspot resistance in sugar beet? (Proof of Concept).

2. The development of progeny from crosses of the PT7#12 transgenic with high quality genotypes C842 and 9933 developed by Bob Lewellen in Salinas, CA.

Materials and Methods:
We had planned to use seeds obtained from the transgenic genotype PT7#12 to develop plants suitable for *Cercospora* leafspot evaluation under controlled environmental conditions in a growth chamber or in the greenhouse. Artificial field inoculation with *Cercospora beticola* and leafspot scoring has previously been used to examine the relative leafspot susceptibility of various genotypes of sugar beet (Kuykendall, unpublished data). However all of the plants developed from seed at Beltsville, MD bolted very early, negating the possibility of obtaining vegetative plants of sufficient maturity and size for *Cercospora* leafspot evaluation. Therefore our seed from greenhouse-grown PT7#12 seed was sent to Lee Panella in Ft. Collins, CO to cross. It was decided to use improved germplasm from R. T. Lewellen with multiple disease resistances. The transgenic plants grown in Ft. Collins are being crossed sugar beet genotypes 'C842' and '9933', both out of Salinas, CA. PT7#12 is being crossed to 31 parents (See Table below) – paired crosses (full sib families) – and each of the transgenic parents is being selfed. There potentially are two more transgenic plants to cross – we are waiting until both the males and females are ready. Meanwhile vegetative plants of sufficient size and maturity for *Cercospora* leafspot evaluation of PT7#12 and its Rel-1 parent are now being successfully developed at Beltsville. Both hybrid and selfed progeny of the crosses of CFP transgenic PT7#12 with the sugar beet genotypes will be returned to Beltsville to determine *cfp* insertion and for *Cercospora* leafspot evaluation.

C842 is released from Salinas – It is rhizomania resistant (RhzmR), monogerm (*mm*), self-fertile (S^f), Curly top resistant (CTR), segregating for genetic male sterility (*A-:aa*), and green hypocotyl color (*R-:rr*), - it is a facilitated random mated population with variable reaction to bolting, Erwinia, and powdery mildew.

9933 comes from 8933, which consists of - #s *aa* x *A*. It is rhizomania resistant (RhzmR), multigerm (*MM*), self-fertile (S^f), Curly top resistant (CTR), Virus yellows resistant (VYR), Powdery mildew resistant (PMR), Erwinia resistant bolting resistant, segregating for genetic male sterility (*A-:aa*), root aphid resistance, and green hypocotyl color (*R-:rr*) w/ normal cytoplasm.

PT7#12, transgenic sugarbeet (*Beta vulgaris* L.), clone 'Rel-1' with the cercosporin toxin export gene, *cfp*, from *Cercospora kikuchi* introduced by D. Kuykendall of the Beltsville plant pathology laboratory

Time Line of Anticipated Accomplishments:

The evaluation of *Cercospora* leafspot resistance in transgenic sugar beet genotypes, relative to parental germplasm tolerance is needed to prove or disprove the concept that the CFP gene can be useful in enhancing leafspot resistance in sugar beet. The progeny from crosses of the PT7#12 transgenic with high quality genotypes C842 and 9933, developed by Bob Lewellen in Salinas, CA, will allow us to evaluate potential *Cercospora* leafspot resistance in the sugar beet genetic background.

The incorporation of novel genes from transgenic sources into agronomically acceptable germplasm can be a long term proposition - results do not occur overnight. This is the type of long-term germplasm development is what ARS is well-suited to perform.

Now that vegetative plants of sufficient size and maturity are being developed from seed of PT7#12 at Beltsville, MD, this will hopefully allow use to do the following:

1) Produce at least a preliminary proof of concept test for the hypothesis that CFP expression in transgenic sugar beets can be used to enhance *Cercospora* leafspot resistance, in as soon a timeframe as late 2005 or early 2006.

2) Produce more viable seed of greater maturity and size---to be used to develop more plants for additional crosses as outlined above.

Research Progress 2004:

Sufficient viable seed of CFP transgenic PT7#12 were produced at Beltsville for a number of crosses made in Ft. Collins, CO with germplasm developed at Salinas, CA. We have 31 crosses of individuals of PT7#12 with genotypes C842 and 9933. Seed have already been harvested from 14 crosses and the remainder are paired and flowering. Evaluations of these crosses will begin in fall of 2005 and results will be reported in next year's BSDF annual report. Plants from these full-sib families populations producing some biennial plants will be vernalized for 120 days and the populations are being increased (i.e., random mated using the genetic male sterility where possible). The annuals will be handled in a similar fashion once the F_1 populations have been increased. Biennial plants will be evaluated for resistance to *Cercospora* leaf spot and the number of 'cfp' alleles determined.

Greenhouse Crosses from January to April 2005 in the Greenhouse at Fort Collins, CO.				
ID[1]	Hypocotyl color	PF[2] or MS	Plant #	Number Assigned to Seed
2004A001	Pink	PF	#1	20041021H-01s
2004A002	Pink	MS	#1	20041021H2-01

Greenhouse Crosses from January to April 2005 in the Greenhouse at Fort Collins, CO.

ID[1]	Hypocotyl color	PF[2] or MS	Plant #	Number Assigned to Seed
2004A001	Pink	PF	#2	20041021H-02s
2004A002	Pink	MS	#2	20041021H2-02
2004A001	Pink	PF	#3	20041021H-03s
2004A002	Pink	MS	#3	20041021H2-03
2004A001	Pink	PF	#4	20041021H-04s
2004A013	Pink	MS	#4	20041021H3-04
2004A001	Green	PF	#5	20041021H-05s
2004A013	Pink	PF	#5	20041021H3-05
2004A001	Pink	PF	#6	20041021H-06
2004A002	Pink	MS	#6	20041021H2-06
2004A001	Pink	PF	#7	First fem. died, no sd;not harv yet
2004A002	Pink	MS	#7	2nd substituted
2004A001	Pink	PF	#9	20041021H-09s
2004A013	Green	MS	#9	20041021H3-09
2004A001	Pink	PF	#10	20041021H-10s
2004A013	Pink	MS	#10	20041021H3-10
2004A001	Pink	PF	#12	harvested - no designation yet
2004A002	Green	PF	#12	harvested - no designation yet
2004A001	Pink	PF	#13	harvested - no designation yet
2004A002	Pink	MS	#13	harvested - no designation yet
2004A001	Green	PF	#14	harvested - no designation yet
2004A002	Pink	MS	#14	harvested - no designation yet
2004A001	Pink	PF	#15	harvested - no designation yet
2004A002	Green	MS	#15	harvested - no designation yet
2004A001	Pink	PF	#16	harvested - no designation yet
2004A002	Green	MS	#16	harvested - no designation yet
2004A001	Pink	PF	#18	harvested - no designation yet
2004A002	Green	PF	#18	harvested - no designation yet
2004A001	Green	PF	#19	harvested - no designation yet
2004A002	Pink	PF	#19	harvested - no designation yet
2004A001	Pink	PF	#20	very little seed
2004A013	Green	PF	#20	harvested - no designation yet
2004A001	Pink	PF	A	paired and flowering
2004A002	Pink	MS	A	paired and flowering
2004A001	Pink	PF	B	paired and flowering
2004A002	Pink	MS	B	paired and flowering
2004A001	Pink	PF	C	paired and flowering
2004A002	Pink	MS	C	paired and flowering
2004A001	Pink	PF	D	paired and flowering
2004A002	Pink	MS	D	paired and flowering

Greenhouse Crosses from January to April 2005 in the Greenhouse at Fort Collins, CO.				
ID[1]	Hypocotyl color	PF[2] or MS	Plant #	Number Assigned to Seed
2004A001	Pink	PF	E	paired and flowering
2004A002	Pink	MS	E	paired and flowering
2004A001	Green	PF	F	paired and flowering
2004A002	Pink	MS	F	paired and flowering
2004A001	Pink	PF	G	paired and flowering
2004A013	Green	PF	G	paired and flowering
2004A001	Pink	PF	H	paired and flowering
2004A013	Green	PF	H	paired and flowering
2004A001	Pink	PF	I	paired and flowering
2004A002	Pink	MS	I	paired and flowering
2004A001	Green	PF	J	paired and flowering
2004A002	Pink	MS	J	paired and flowering
2004A001	Pink	PF	K	paired and flowering
2004A002	Green	PF	K	paired and flowering
2004A001	Pink	PF	L	paired and flowering
2004A002	Green	PF	L	paired and flowering
2004A001	Green	PF	M	paired and flowering
2004A002	Pink	MS	M	paired and flowering
2004A001	Pink	PF	N	paired and flowering
2004A002	Pink	MS	N	paired and flowering

[1]2004A001 = PT7#12, transgenic sugarbeet (*Beta vulgaris* L.), clone 'Rel-1' with the cercosporin toxin export gene, *cfp*, from *Cercospora kikuchi* introduced by D. Kuykendall of the Beltsville plant pathology laboratory.
2004A002 = 'C842' sugarbeet (*Beta vulgaris* L.) germplasm, a biennial, disease resistant germplasm developed by R.T. Lewellen (USDA-ARS, Salinas, CA).
2004A013 = '9933' sugarbeet (*Beta vulgaris* L.) Germplasm, a biennial, disease resistant germplasm developed by R.T. Lewellen (USDA-ARS, Salinas, CA).
[2]PF = Pollen Fertile, and MS = genetic male sterile (*aa*).

SUGARBEET RESEARCH

2004 REPORT

Section F

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Dr. Charlie Rush, Professor of Plant Pathology

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Texas Agricultural Experiment Station
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Mutation and Genetic Variability among Isolates of BNYVV

C.M. Rush, D. Jones, and K. Steddom

Texas Agricultural Experiment Station, 2301 Experiment Station Rd., Bushland, TX 79012

Historically, *Beet necrotic yellow vein virus* (BNYVV), which causes the disease rhizomania, has caused major reductions in root yield and quality where ever it occurred. In the United States, the disease was first identified in California but now occurs in all major sugar beet production regions of the country. Fortunately, strong genetic resistance, conferred by the *Rz* gene, has been incorporated into regionally adapted cultivars, allowing profitable sugar beet production in areas infested with the pathogen. However, in 2002, plants in a field in the Imperial Valley of California that was planted to a rhizomania tolerant cultivar began to express symptoms of rhizomania. Large strips of diseased plants occurred across the field and it was soon verified that BNYVV had overcome genetic resistance. In other regions of the US, individual plants in fields planted to rhizomania tolerant cultivars have also become infected by BNYVV and developed diagnostic symptoms of severe rhizomania. These individual symptomatic plants in fields of apparently healthy plants are called "blinkers". Although patterns of disease development have varied between California fields and those in other production regions in the US, the breakdown of genetic resistance has caused considerable concern among those involved in sugar beet production.

In the Imperial Valley of California, soil samples were taken in fields planted to rhizomania tolerant cultivars from areas exhibiting typical symptoms of rhizomania. BNYVV was baited from the soil using rhizomania resistant plants and then the virus was extracted and purified from the infected bait plants. Genetic analysis of the Imperial Valley strain of BNYVV (CIV-BNYVV) indicated that it was 99% identical to standard isolates of BNYVV that cannot overcome the *Rz* gene. This suggested that the ability of CIV-BNYVV to overcome the *Rz* gene was the result of a minor genetic change in the virus and not a major shift in genetic make up due to recombination, reassortment, or a major deletion. Because of the natural variability among isolates of plant RNA viruses, identification of the specific mutation that allows CIV-BNYVV to overcome the *Rz* gene will be difficult. However, Dr. Hsing Yeh Liu with USDA-ARS in Salinas, CA discovered specific sequences in CIV-BNYVV that appear to be unique and may facilitate identification of this strain from soil samples.

In Minnesota, in fields planted to rhizomania tolerant cultivars, disease primarily has been observed on individual plants and not in large strips or spots in the field. It is well known that in the increase of genetically resistant seed there is always a low percentage of seed that, for various reasons, do not possess the gene that confers the resistance. It is also recognized that there are a number of other reasons that could account for disease development in individual plants. Therefore, the primary question was whether the individual symptomatic diseased plants, i.e., blinkers, actually possessed the *Rz* gene.

Materials and Methods

Blinkers and apparently healthy beets were collected from three strip trials in Minnesota near, Crookston, Morehead, and Willmar. At each field location, a minimum of eight blinkers and two apparently healthy beets were collected from each cultivar represented in the strip trial. Rhizomania tolerant cultivars included in the test included Beta 1305 and 4818, Crystal 826,

Hilleshog 2411, 2463, 2467, and 2469, Seedex 0831 and Rezult, and Vanderhave 46177 and 46519. Beta 3800 and Crystal 725 were included as susceptible controls. Each individual beet was rated for rhizomania severity on a 0 – 4 scale, with 0 = healthy, no symptoms and 4 = severe rhizomania symptoms, such as stunting, constriction and proliferation of lateral roots, and roots with fungal disease symptoms were discarded. Leaf chlorosis was quantified using an integrating sphere hyperspectral radiometer and leaf, root and rhizosphere soil samples were collected and set to the plant pathology laboratory in Bushland. Root samples were tested by DAS-ELISA for presence of BNYVV and, after freeze-drying, leaf samples were tested for presence or absence of the *Rz* gene. Percent sucrose was determined for blinkers and apparently healthy beets for each cultivar collected at each field location.

Results and Discussion

The rhizomania strip trials used in this study provided an ideal location for collecting samples from a large number of rhizomania tolerant cultivars. Sites for the strip trials were initially selected because of anticipated heavy, relatively uniform disease pressure. Disease was most severe at Morehead and least at Willmar, but disease incidence was adequate at each site and replication of each cultivar in the test provided easy access to blinkers.

Use of radiometry to quantify leaf chlorosis proved to be an effective technique (Figure 1). Plotting reflectance at wavelengths from 400 – 700 nm revealed that differences between apparently healthy beets and blinkers were best observed at 555nm. At this wavelength, blinkers displayed significantly higher percent reflectance than healthy beets but differences between blinkers with and without the *Rz* gene were not always significant.
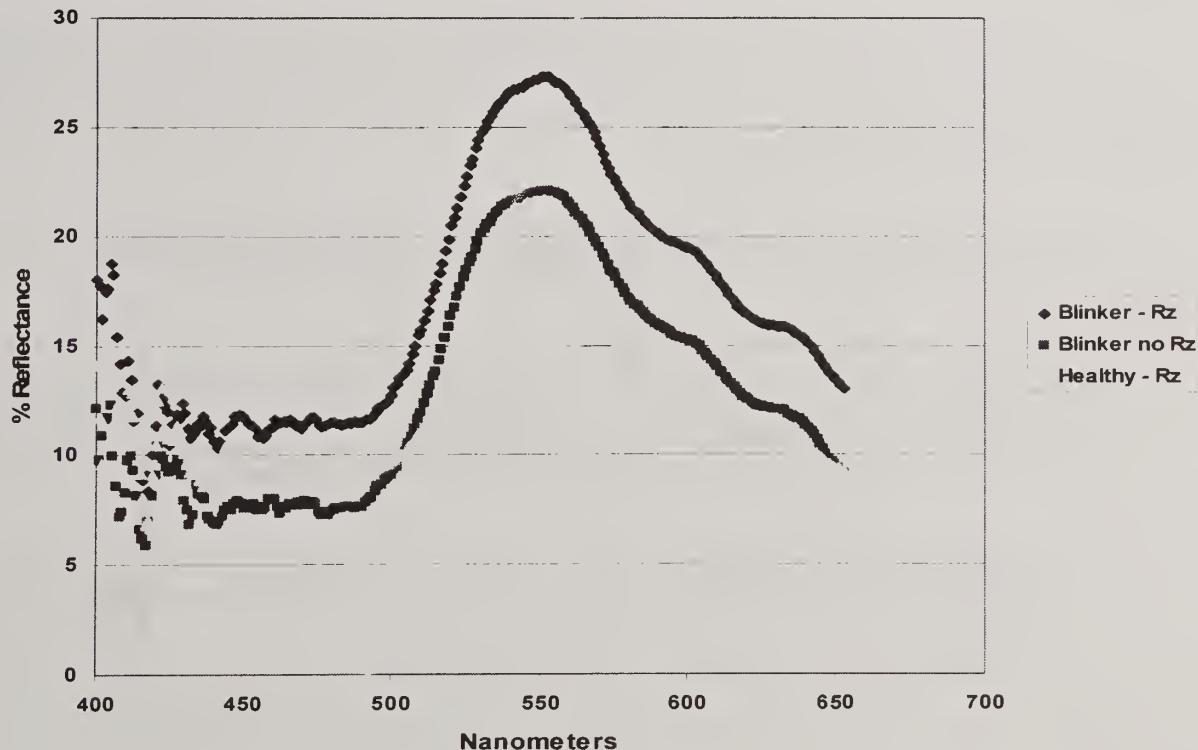

Figure 1. Percent reflectance from leaf tissue obtained from blinkers and healthy beets.

Differences between apparently healthy beets and blinkers were, for the most part, as expected (Table 1). A significantly greater percentage of healthy plants tested positive for the *Rz* gene, compared to the blinkers, but a significantly lower percentage of the healthy plants tested positive for BNYVV in the ELISA test. Blinkers had a significantly higher disease rating and higher reflectance readings at 555nm than the apparently healthy beets and also greatly reduced sucrose content.

Table 1. Comparisons between healthy and blinkers for several variables.

Plant Type	Disease Rating	%Positive for *Rz* Gene	Reflect. 555 nm	%Positive ELISA	Sucrose
Blinkers	2.95	52%	26.58	88%	13.98
Healthy	1.14**	80%**	19.04**	44%**	15.67 **

However, when comparing disease ratings between blinkers with or without the *Rz* gene, there was no difference (Table 2). This indicated that presence of the *Rz* gene in these plants was not conferring any resistance to BNYVV and was another indication that the virus has actually mutated into a new virulent strain.

Table 2. Comparisons between blinkers with and without the *Rz* gene.

Rz Category of Blinkers	% *Rz* Gene in Blinkers	Disease Rating	%Positive ELISA	Reflectance at 555 nm
Rz Negative	57%	2.88	95%	27.28
Rz Positive	42%*	2.93 ns	80%*	27.61ns

The fact that 42 percent of the blinkers we sampled possessed the *Rz* gene but it had no beneficial effect in reducing disease severity is worrisome. However, the overall incidence of

blinkers in the field at each test site was < 5%. Still, if the blinkers that possessed the *Rz* gene were the result of infection by a new strain of BNYVV, capable of overcoming resistance, the incidence of rhizomania will be much higher in the next crop. However, the results from this study are inconclusive and do not prove the existence of a new virulent strain. It is well known that several factors other than mutation of the pathogen, such as high inoculum density or the combination of minor genes, can impact severity of rhizomania. Additional study is needed to determine the etiology of blinkers in Minnesota sugar beet production areas.

SUGARBEET RESEARCH

2004 REPORT

Section G

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Table of Contents

Colorado State University
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Progress Report
BEET SUGAR DEVELOPMENT FOUNDATION
FY 2004

Project Title: New Strategies for Modifying Sucrose Distribution in Sugarbeet

Project Number: 840

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                  Fort Collins CO 80523

Other Personnel Involved: graduate student

Project Location: Biology Department
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Justification of Research:

Sucrose accumulated in the sugar beet tap root is synthesized in the leaf and then transported to the root in the phloem cells of the plant's vascular system. The proton-coupled sucrose transport protein mediates the key step in the long-distance transport of newly synthesized sucrose from the leaf to the taproot because it is responsible for sucrose accumulation into the leaf phloem cells and that activity drives sucrose flux to the tap root. We recently discovered a control pathway that regulates the activity of the sucrose transporter and, because of the transporter's role in loading the phloem, this regulatory system appears to control sucrose export from the leaf (Chiou and Bush 1998, Bush 1999). This was a very significant finding because loading the vascular system for sucrose export from the leaf determines how much sucrose is delivered to the tap root. Defining the biochemical and molecular steps involved in controlling sucrose delivery to the beet will allow us to develop new strategies for manipulating productivity.

Recent Progress

Research this year focused on two areas: 1) experiments aimed at defining the key steps in sucrose-sensing regulatory pathway described above and 2) a biotech approach to express a hyperactive form of the sucrose transporter in the leaf phloem with the goal of increasing the amount of sucrose transported to the storage beet. Advances this year on sucrose sensing included identifying mutant plants that are not sensitive to sucrose. We are now using genetic strategies to identify the mutated genes, which we hope are components of the sensing pathway. We are also developing a novel method to determine all the genes expressed in the plant's vascular cells. This will aid our efforts to understand the sucrose-dependent regulation of sucrose transport. For objective two, we are collaborating with Marc Lefebvre (Advanta Biotechnology) to make transgenic plants expressing the hyperactive transporter in the leaf phloem cells. We are constructing the expression vector and will send that to Marc for beet transformation. Once transgenic plants are produced, my lab will examine their growth and the impact of the hyperactive transporter on sucrose accumulation in the beet. Five manuscripts have

been published reporting these results and summarizing the status of the field (Bush and Coruzzi, 2000; Vaughn, Harrington, & Bush, 2002, Ransom-Hodgkins et al. 2003, Harrington and Bush 2003, and Bush 2004).

Publications resulting from BSDF support

Coruzzi G and Bush DR 2001. Nitrogen and carbon nutrient and metabolite signaling in plants. *Plant Physiol* 125: 65-68

Vaughn MW, Gregory N. Harrington, and DR Bush 2002. Sucrose-mediated transcriptional regulation of sucrose symporter activity in the phloem. *Proc. Natl. Acad. Sci. USA* 99:10876-10880

Ransom-Hodgkins W, MW Vaughn, and DR Bush 2003. Protein phosphorylation mediates a key step in sucrose-regulation of the expression and transport activity of a beet proton-sucrose symporter. *Planta* 217:483-489

Harrington GN and Bush DR 2003. The bifunctional role of hexokinase in metabolism and glucose signaling. *Plant Cell* 15: 2493-2496

Bush DR 2004. Functional analysis of proton-coupled sucrose transport. *In:* Membrane Transport in Plants. *Ed.* Michael Blatt. p. 135-147 Blackwell Publishing

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Chiou TJ and DR Bush 1998. Sucrose is a signal molecule in assimilate partitioning. Proceedings of the National Academy of Sciences USA 95:4784-4788

Bush DR 1999. Sugar transporters in plant biology. Current Opinion in Plant Biology 2:187-191

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